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DENITRIFICATION AND TILLAGE  
RELATIONSHIPS ON TWO  
EASTERN SOUTH DAKOTA SOILS

by

Theresa Helen Lemme

A dissertation submitted

in partial fulfillment of the requirements for the

degree of Doctor of Philosophy

Major in Agronomy

South Dakota State University

1988

DENITRIFICATION AND TILLAGE

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EASTERN SOUTH DAKOTA SOILS

EASTERN SOUTH DAKOTA SOILS

This dissertation is approved as a creditable and independent investigation by a candidate for the degree, Doctor of Philosophy, and is acceptable for meeting the thesis requirements for that degree. Acceptance of this thesis does not imply that the conclusions reached by the candidate are necessarily the conclusions of the major department.

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# DENITRIFICATION AND TILLAGE

## RELATIONSHIPS ON TWO EASTERN SOUTH DAKOTA SOILS

### Abstract

Theresa H. Lemme

Tillage practices have been shown to affect denitrification. Denitrification is an important process involving the loss of nitrogen from agricultural soils. This study investigates denitrification on a Beadle clay loam and a Worthing silty clay loam in a landscape in Eastern South Dakota. Comparisons were made in 1986 and 1987 of these soils under four different tillage systems: Moldboard plowed, chisel plowed and disked, ridge tillage, and no tillage systems. Soil core samples (0 to 5 cm) from continuous corn plots were utilized to determine nitrogen loss by denitrification and volumetric moisture biweekly through the growing seasons. In addition, organic matter residue amount, bulk density, and precipitation were also reported.

In the laboratory, glucose and nitrate additions were related to denitrification in the Worthing soil of different aggregate sizes. Glucose and nitrate additions stimulated denitrification when added at 625-ug glucose g<sup>-1</sup> soil. However, amounts in excess of this failed to stimulate



activity.

Five sizes of Worthing soil aggregates (<1, 1 to 2, 2 to 4.8, 4.8 to 6.4, and 6.4 to 9.5 mm) were maintained at 45% volumetric moisture on a sand tension table and subsequent denitrification rates were measured. No differences were seen.

Tillage practices were found to relate to denitrification rates. No differences were observed between the other three tillage treatments, however the Worthing lowland soil better potential to denitrify than the Beadle upland soils relating to the higher volumetric moisture on the Worthing soil. Estimated denitrification for the two years varied between 3- and 36-kg nitrogen loss  $\text{ha}^{-1} \text{yr}^{-1}$  on the Beadle soil, and 17- to 36-kg nitrogen loss  $\text{ha}^{-1} \text{yr}^{-1}$  on the Worthing soil.

A computer model was developed to predict denitrification rates as a function of volumetric moisture. A volumetric moisture value of 22.3% or less would completely inhibit denitrification. Soil volumetric moistures found in this study varied between 15 and 55%. Of the parameters measured in this study, volumetric moisture appears to be the most significant factor. However, potential losses of nitrogen by denitrification should be considered in designing soil management schemes since tillage methods can greatly control soil moisture.

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My parents and siblings have helped shape me and I want to acknowledge that sharing spirit which runs through all of them. With the knowledge that they will always be there, I have the courage to try.

My dearest husband, Gary, and son, Carl, deserve so much for supporting me through the long trek. Each has made many sacrifices so that I might continue. Both of them deserve all the love I can give them.

The Lord has made each of us uniquely. I hope and pray that I might live up to His plans for me.

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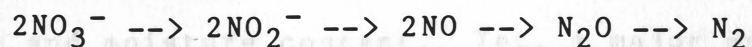
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## INTRODUCTION

The availability of nitrogen to plants is considered a major factor limiting production. The balance that exists between nitrogen fixation, nitrogen mineralization, and denitrification along with fertilizer inputs, determines the amount of nitrogen available to plants. On a worldwide basis, the process of nitrogen fixation provides more available nitrogen than mineralization or chemical nitrogen fertilizer formation. Fixation of atmospheric nitrogen can be carried out symbiotically, as in the legume-rhizobium symbiotic relationship, or nonsymbiotically by free living microorganisms such as Azotobacter. This microbial process can account for the addition of 61- to 112-kg N ha<sup>-1</sup> year<sup>-1</sup> in South Dakota soils. Soil organic matter and crop residue nitrogen is mineralized as it is decomposed by aerobic heterotrophic bacteria, actinomycetes, and fungi to release ammonium ions which are quickly oxidized to the nitrite and nitrate form by species of Nitrosomonas, Nitrobacter and related genera. (1980a) has reported on

Nitrogen is lost from the soil ecosystem by plant utilization, denitrification, leaching through the profile and soil erosion. Denitrification involves the reduction of nitrates to nitrogen gas or nitrogen oxides which can be lost through diffusion to the atmosphere. This process can be carried out by various soil microorganisms (facultative

anaerobes) which utilize the nitrates as terminal electron acceptors by the metabolic pathway outlined below:



Microorganisms use nitrate ( $\text{NO}_3^-$ ) as an electron acceptor with the result that nitrite ( $\text{NO}_2^-$ ) and nitric oxide ( $\text{NO}$ ); and then nitrous oxide ( $\text{N}_2\text{O}$ ) and dinitrogen ( $\text{N}_2$ ) gases are produced (Colbourn, 1984). Payne (1981) considers that denitrification ranks with nitrogen fixation and photosynthesis in importance to agronomy. It has been estimated that up to 86% of applied nitrogen fertilizer may be lost through denitrification. Loss depends upon crop, soil type, soil moisture, temperature, and tillage (Bremner and Shaw, 1958b; Duxbury et al., 1982; Firestone, 1982; Klemmedtsson et al., 1977; Rolston, 1976, 1979; Craswell, 1978; Kissell, 1978). Nitrogen deficiency by denitrification can cause lower crop yields and therefore reduce profitability.

How soil microbial populations are impacted by the amount of crop residue remaining in the soil is little understood. Doran (1980a) has reported an increase in numbers of nitrifying bacteria, denitrifying bacteria, actinomycetes, fungi and other bacteria in plots receiving surface applied corn stover. The tillage practice used determines the residue cover found in a particular farming system. Conservation tillage is a planting and tillage method resulting in a crop residue cover of at least 30%

(USDA, 1984). This can benefit the farmer by decreasing erosion and soil compaction while increasing soil organic matter and moisture content. Yet, a major question is, will this increase in organic matter and moisture also increase denitrification and potentially result in a significant loss of available nitrogen? Indications are that significant loss of  $\text{NO}_3^-$  could occur in soil receiving no-till and reduced tillage treatments compared to a conventionally tilled soil (Doran, 1980a and 1980b; Rice and Smith, 1982), yet, no difference in nitrogen loss among tillage systems was detected on poorly drained soils of Maryland and West Virginia (Staley, 1982).

Most farms in eastern South Dakota include both well and poorly drained soils. Conservation tillage systems are rapidly being adopted by producers in South Dakota. It has been estimated that 65% of the United States major crops (corn, soybeans, sorghum, wheat, oats, barley, and rye) will be grown using a no-tillage cropping system by the year 2000 and up to 78% in the year 2010 (Phillips et al., 1980).

The purpose of this study is to determine some of the parameters controlling denitrification on two soils under a continuous corn crop within a landscape. Some of the parameters studied were soil moisture, soil glucose content, tillage method, percent residue cover, and soil aggregate size. A better understanding of the magnitude of

nitrogen losses by denitrification within various tillage systems under both well and poorly drained conditions will not only add to our understanding of the structure and function of these systems, but will provide producers with information needed when considering tillage options.

The objectives of this study were:

1. To study the effect of tillage systems upon soil denitrification on well drained Beadle and poorly drained Worthing soils.
2. To study the effect of soil moisture on soil denitrification under continuous corn plots.
3. To study the effects of drainage and tillage upon extractable carbohydrate calculated as 'glucose equivalents' (pre and post mid-season cultivation).
4. To study the effect of aggregate size and moisture tension on denitrification.
5. To study the effect of glucose amendments on denitrification in soil of known aggregate size.



therefore indirectly LITERATURE REVIEW are not have other

I. DEFINITION OF DENITRIFICATION Soil for Agricultural

Denitrification is the biological process through which nitrate-N is returned to the atmosphere as nitrogen gas or oxides of nitrogen. It is defined in the Soil Science Society of America glossary of soil science terms (1979) as "the microbial reduction of nitrate or nitrite to gaseous nitrogen either as molecular nitrogen or as an oxide of nitrogen". Most microbiologists expand on this definition by designating nitrogen oxides as terminal electron acceptors for respiratory electron transport leading to a more reduced nitrous oxide. Energy is conserved by electron transport phosphorylation. Denitrification is a poorly understood process within the realm of nitrogen transformations. Estimates of agricultural nitrogen loss through denitrification range from 0 to 70% of applied fertilizer nitrogen (Craswell, 1978).

Diffusion of nitrous oxide from the soil has generated a large interest in denitrification from an ecological viewpoint. Nitrous oxide has been pinpointed as one of the compounds which break down the ozone mantle surrounding the earth (Crutzen, 1970; Crutzen and Ehhalt, 1977; Johnston, 1971). A global increase in  $N_2O$  would decrease the ozone mantle, increasing the ultraviolet light reaching the earth (Boyd, 1984; Yung et al., 1976), and

therefore indirectly increase skin cancers and have other deleterious effects on health (Council for Agricultural Science and Technology, 1976). Rosswall (1976) suggests that by plowing and cultivating our prairies, we have changed the ecosystem. With an increased rate of organic matter mineralization and higher nitrate fertilizer use, denitrification occurs at an accelerated rate. Rosswall believes that because of the size of the nitrogen molecule, nitrous oxide could have a more intense greenhouse effect than does carbon dioxide, the more commonly studied gas.

## II. DENITRIFICATION MEASUREMENTS

There are a number of published methods of measuring denitrification. These include  $^{15}\text{N}$  tracer methods and soil cover techniques in the field (Payne and Dowdell, 1984) and soil slurry and soil core techniques in the laboratory (Ryden, 1979; Hilton, 1985; Bonrud, 1985). Each has its limitations and benefits, as will be discussed below.

### A. USE OF $^{15}\text{N}$ TRACER METHODS

Measuring denitrification by difference in the field is accomplished by the use of  $^{15}\text{N}$  labelled fertilizer. Inputs and outputs of  $^{15}\text{N}$  are compared and it is assumed that any unrecovered  $^{15}\text{N}$  is loss by denitrification. A tracer method using  $^{13}\text{N}$  allows observation of atoms at various points along the denitrification pathway (Payne and



Dowdell, 1984). This technique proves to be expensive to use and the  $^{13}\text{N}$  has a short half life.

#### B. USE OF ACETYLENE BLOCKAGE

Ryden et al. (1979) and Smith et al. (1978) have confirmed that measurement of nitrous oxide production in the presence of acetylene is a valid estimator of total gaseous nitrogen which would be produced without the presence of acetylene. Acetylene inhibits the  $\text{N}_2\text{O}$  reductase enzyme, thus preventing  $\text{N}_2\text{O}$  reduction to nitrogen gas ( $\text{N}_2$ ). Acetylene blockage has developed as the method of choice in measuring denitrification (Yoshinari et al., 1977; Smith et al., 1978; Ryden et al., 1979). The quantity of  $\text{N}_2\text{O}$  produced can be easily determined using a gas chromatograph and is a valid measure of the total gaseous  $\text{N}_2$  which would have been produced if acetylene were omitted (Ryden et al., 1979; Smith et al., 1978). This confirmation allows scientists to accurately measure denitrification from soil samples by a variety of methods.

#### C. USE OF SOIL COVERS OR CHAMBERS

In situ soil covers or chambers have been used to collect the  $\text{N}_2\text{O}$  evolved after acetylene addition to the soil to block  $\text{N}_2\text{O}$  reduction to  $\text{N}_2$  (Ryden, 1979; Hilton, 1985). A cover is placed over the soil and acetylene is added through metal tubes inserted below the surface of the soil. Yoshinari et al. (1977) and Balderston et al. (1976)

determined that acetylene presence would block reduction of  $N_2O$  to  $N_2$  in cultures of denitrifying organisms. The acetylene is allowed to perfuse the soil and  $N_2O$  diffusing from the soil surface is collected by the cover. The gases are drawn from the cover and trapped on a selective adsorbant. The adsorbant is taken to the laboratory where the  $N_2O$  gas is chemically removed and can be quantitatively analyzed by gas chromatography. Another chamber method involves setting the chamber on the soil surface and measuring  $N_2O$  emission under natural conditions (no acetylene addition) (Matthias et al., 1980 and Denmead, 1979). Denmead (1979) devised a system where diffusing gases would be drawn directly into an infrared gas analyzer. The advantages of this technique are that the soil is not disturbed, both  $N_2O$  emission and a sink activity can be monitored and no special equipment is needed. The disadvantage of cover techniques is that the equipment is bulky and only single soil samples can be analyzed at a time.

#### D. USE OF SOIL SLURRIES

Another method of analysis involves mixing water with a soil sample to create a soil slurry. The slurry is prepared in a gas-tight container to which acetylene is added. The soil slurry mixture is then incubated prior to analysis of the gas in the container headspace by gas

chromatography (Betlach and Tiedje, 1981). This induces optimum conditions for denitrification and does not emulate natural conditions.

#### E. USE OF SOIL CORE TECHNIQUES

In vitro soil core techniques have also been used to estimate denitrification (Stout, 1984). A soil core is removed to an airtight container and transported to the lab. Acetylene is added to the soil cores in their airtight container to inhibit  $N_2O$  reduction (Aulakh, 1982; Rice and Smith, 1982; Parkin, 1984; Bonrud, 1985). After incubation,  $N_2O$  gas content of the airspace in the container can be analyzed by a gas chromatograph. Soil core methods have become useful as a rapid denitrification assay. Numerous small cores can be collected and transported to the laboratory for acetylene treatment, incubation, and analysis with a gas chromatograph equipped with an electron capture device (Groffman, 1985; Rice and Smith, 1982). A disadvantage of this method is that the soil is disturbed and not under natural conditions. An advantage is that the disturbance can be kept to a minimum and multiple soil samples can be analyzed quickly and simultaneously. A study by Bonrud (1985) compared results from a chamber method and a core method of denitrification determination and found a significant correlation between these methods, thus supporting a concept of interchangeability of data.

### III. FACTORS AFFECTING DENITRIFICATION

Nitrogen losses by means of denitrification are apparently increased by any factor which decreases the aerobic sites in the soil. Soil texture, soil moisture content, crop cover, tillage, bacterial numbers, residues, carbon source, temperature and pH also play some part in the complex picture by controlling the driving forces of denitrification. Factors which decrease the aerobic sites include an increase in moisture content, whether by tillage method rainfall, or irrigation, and an increase in bulk density. As nutrients in the soil become available through organic matter mineralization, microbial nutritional needs are met.

#### A. MOISTURE

An increase in moisture fills pore spaces in the soil and results in lower oxygen concentration which will be discussed later. The denitrifying bacteria reduce nitrate or nitrite to nitrogen gas or nitrous oxide in the absence of oxygen (Firestone, 1982). In Michigan, moisture increase has been shown to result in a rapid increase in denitrification rate on a sandy loam soil after a 6 hour lag period (Smith and Tiedje, 1979). Water filled pore space above 60% was found to decrease aerobic microbial activity and to increase denitrification activity (Linn and Doran,



1984b and Bremner and Shaw, 1958b). Aulakh et al. (1984) performed a multiple regression analysis that showed that volumetric water content, temperature, inorganic nitrate-N and ammonia-N did account for 37 to 66% of the variation in denitrification rate. Bulk density values were included in the regression but were not a significant variable. Bremner and Shaw (1958a) demonstrated that denitrification did not occur in moist soils when the soil was aerated continuously while carbon and nitrogen were added, thus showing that the degree of aeration limited the ability of the microorganisms to denitrify. In undisturbed soils, denitrification activity may increase significantly under saturated conditions or when large aggregates are present because the water acts as a barrier to oxygen diffusion. Mean rates of denitrification were found to exceed  $0.2\text{ kg N ha}^{-1}\text{ day}^{-1}$  only when a loam soil had over 20% moisture (w/w) and had over five micrograms nitrate-N  $\text{g}^{-1}$  in the upper 20 cm (Ryden, 1983). Soil temperatures had to exceed 5 to 8 °C to initiate denitrification.

#### B. AGGREGATE SIZE

Denitrification activity can be found in soils with atmospheric oxygen concentrations in the macropore space (Currie, 1986). Greenwood and Goodman (1967) measured oxygen diffusion into soil crumbs and found that oxygen content decreased rapidly with depth under the crumb

surface. Oxygen diffusion into soil aggregates affects the amount of denitrification which can occur, yet there is evidence that denitrification enzymes are produced when sufficient oxygen is present to inhibit enzyme activity (Cho, 1982). Aggregated unsaturated soils contain anaerobic areas and if nitrate and decomposable organic matter are also present, denitrification can occur in those anaerobic zones (Leffelaar, 1986). Sexstone et al. (1985b) studied oxygen diffusion through soil aggregates. Some aggregates even though they contained anaerobic centers did not denitrify. They found that aggregates of 8 mm diameter could exhibit anaerobic centers while larger aggregates, collected from a native prairie soil might exhibit small or no anaerobic centers. Myrold and Tiedje (1985) found that in undisturbed soil, or those saturated with water, denitrification capacity may increase. This indicates that anaerobic sites are not the only factor involved in denitrification rate.

### C. SOIL TEXTURE

Denitrification losses on a sandy loam and a clay loam soil as determined by Sexstone et al. (1985a) were shown to be most influenced by texture and moisture. The lag periods associated with the two soils varied. Maximum denitrification occurred within three to five hours after 7 cm of water was added to a sandy loam profile. Denitri-

fication returned to pre-moisture rates within 12 hours. The clay loam profile required eight to twelve hours to reach maximum denitrification levels after wetting with two cm water. Denitrification levels did not return to  $\text{NO}_3^-$  pre-wetting levels until 48 hours later. The air filled porosities of both soils were  $0.37 \text{ m}^3 \text{ m}^{-3}$  after wetting. Nitrogen losses were two times as much in the clay loam soil. They indicate that more nitrogen is lost from the clay loam soil because of the longer duration of enhanced denitrification rate and postulated that a slower water infiltration in the clay loam soil caused this. They believed that with a rainfall over one cm, 38 to 55% of the total nitrogen loss in the late spring would occur within 48 hrs. Westerman and Tucker (1979) determined denitrification by the difference between added soil nitrogen and remaining soil nitrogen forms after field incubation for three, six, nine, and twelve months. They found that most of the losses occurred within the three month period of measureable rainfall on these Sonoran desert sandy loam soils. Total losses varied from 19 to 94% of inorganic nitrogen added to these soils and the highest losses occurred with  $\text{K}^{15}\text{NO}_3\text{-N}$  fertilizer use. Wheat straw additions stimulated immobilization and reduced denitrification losses to 30 to 86% of the nitrogen applied. The smallest losses occurred when  $(^{15}\text{NH}_4)_2\text{SO}_4\text{-N}$  was applied. They conclude that

denitrification is an integral and important part of nitrogen flow through a desert ecosystem. Therefore, significant nitrogen loss can occur depending on soil texture in response to precipitation events, as long as  $\text{NO}_3^-$  or carbon were not limiting.

#### D. CROP TYPE

The crop grown in the soil has been shown to affect denitrification by that soil. Summer fallowed ground had 300% higher denitrification losses than soil with a wheat crop ( $3\text{-kg N ha}^{-1}$  or less) for May through November in Saskatchewan (Aulakh et al., 1982). In a further study, soils with a wheat-fallow rotation lost 2 to 5 times as much N as soils under continuous wheat (Aulakh et al., 1983). The authors believe that both moisture and nitrate-N accumulation during summer fallow directly related to the increased N loss by denitrification.

Sexstone et al. (1985a) and Rolston (1982) report that denitrification response ceased with subsequent rainfall events suggesting that  $\text{NO}_3^-$  may be removed from the active sites by leaching. Firestone (1982) summarized several studies and found that profile characteristics impeding water flow would increase denitrification potential.

#### E. TILLAGE

Tillage has been shown to affect denitrification.



Linn and Doran (1984) show that in the surface 7.5 cm, bulk density, volumetric water, water soluble carbon, organic carbon, and nitrogen content and water filled pore space were greater in no till soils than in conventionally tilled soils. Populations of aerobic and anaerobic microorganisms were also greater in the surface 7.5 cm of no till soils than in conventionally tilled soils. Denitrification activity was greater in no-till than in plowed Maury silt loam (Typic Paleudalfs) in Kentucky (Rice and Smith, 1982) and in a no-till managed Poinsett silty clay loam (Hilton, 1985). Enhanced denitrification activity may cause a lower soil nitrate concentration. Higher nitrogen fertilizer requirements were tentatively attributed to this enhanced activity. In contrast to these results, a study by Carter and Rennie (1982) found no change in total soil organic carbon or nitrogen between tillage systems on four locations under zero and conventional tillage practices for 2, 4, 12, and 16 years. Potential net mineralizable carbon and nitrogen were greater in the surface soil under zero tillage than in soil under conventional tillage. Changes in soils due to tillage vary greatly.

#### F. CARBON AND NITROGEN

Dick (1983) has shown that a no tillage treatment will result in higher organic carbon and nitrogen in the surface soil and less organic carbon and nitrogen lower in

the profile than does the minimum tillage or conventional tillage treatment. Organic carbon decreased twelve to fourteen percent with long term minimum tillage or conventional tillage on a Hoytville soil and decreased 23 to 25% on a Wooster soil. From the surface to a depth of 7.5 cm, a no till soil contained more potentially mineralizable nitrogen than did plowed soils in a study of soils from seven U.S. locations (Doran, 1980b). These studies have all found that no-till systems increase the mineralizable soil nitrogen and therefore the potential for denitrification. Hilton (1985) found that denitrification in a Poinsett silty clay loam appeared to be inhibited by injecting  $\text{NH}_4^+$  in the no-till plots. Another factor found to influence denitrification was the presence of  $\text{NO}_2^-$  which caused a decrease of  $\text{N}_2\text{O}$  reduction to  $\text{N}_2$  (Firestone, 1979; Ryzova, 1979). As the level of nitrate increased in soil, denitrification was found to increase in a number of studies (Betlach and Tiedje, 1981; Firestone, 1982; Ryden, 1983; Ryzhova, 1979), and Stanford et al. (1975a) found that denitrification followed first order kinetics at nitrate concentrations of less than  $40 \text{ mg N L}^{-1}$ .

#### G. BACTERIAL POPULATIONS

Previous work on bacterial populations associated with reduced tillage showed that denitrifiers were present in reduced till soils at 7.31 times the population of those

found in plowed soils (Doran, 1980b). Almost all denitrifying organisms are facultatively anaerobic, capable of anaerobic growth only when nitrogen oxides are present as electron acceptors. Denitrifying organisms can be found throughout the soil and are viable under many conditions. Bremner and Shaw (1958a) determined that the organisms are not detrimentally affected by air-drying or storage of the soil, which explains why the organisms are able to denitrify even after long dry periods in the field soil. Gamble et al. (1977) determined that a diverse group of gram negative chemoheterotrophic motile bacteria were responsible for denitrification in their nineteen studied soils and sediments collected worldwide. Pseudomonas fluorescens and Alcaligenes sp. made up a large portion of their 146 isolates. These two species were also responsible for denitrification in soils studied by Payne (1981) and Colbourn and Dowdell (1984); and Firestone (1982) also includes Agrobacterium, Azospirillum, Bacillus, Rhizobium, and others. McCalla et al. (1962) found that soil microorganisms in subtilled land were more numerous in the top 2.5 cm than in plowed land. Linn and Doran (1984a) in characterizing six surface soils in Illinois, Kentucky, Minnesota, and Nebraska found that the top 7.5 cm of the no till soil system contained 1.27 to 1.31 times the number of anaerobic bacteria found in plowed soils. Denitrification

potential was also greater in these top 7.5 cm than lower in the profile (7.5 to 30 cm). Populations of denitrifiers decrease with increasing depth in a profile (Brar, 1978). Denitrification potential increased lower in the profile in a conventionally tilled soil (Linn and Doran, 1984a). Loss of nitrate nitrogen per bacterial cell varied between 2 and 8 pg in organic matter or sewage amended Windsor sandy loam (Jacobson and Alexander, 1980). Straw, nitrogen additions, and a combination of straw and nitrogen additions produced three times the amount of denitrification in a zero tilled soil compared to a conventionally tilled soil (Aulakh, 1984). In comparing the zero tilled to the conventionally tilled soil with no amendments, twice the amount of denitrification endproducts were seen in zero tilled soil. Four soluble carbon compounds and two plant residues (alfalfa and straw) were used as soil amendments by deCatanzaro and Beauchamp (1985). A rapid rate of denitrification occurred with the soluble carbon compounds and a fairly rapid rate with alfalfa amendment. Straw did not produce the swift rise in denitrification seen with the soluble carbon or the alfalfa amendments.

Presence of a utilizable carbon source is a major factor influencing denitrification rates. Carbon in ample supply can facilitate rapid oxygen consumption, thus creating anaerobic sites in the soil and enhancing



denitrification potential. The most abundant denitrifying organisms are heterotrophic and require organic matter and residue breakdown products. Organic compounds are used as electron donors and as sources of cellular carbon (Firestone, 1982). As more carbon becomes available to the microorganisms, denitrification increases (Firestone, 1982; Koskinen and Keeney, 1982; Letey et al., 1980; Limmer and Steele, 1982; Stanford et al., 1975b; Staley, 1982). This varies with the type of carbon source added. Addition of a readily utilizable energy source, such as glucose, will result in a more rapid denitrification rate. Parkin (1987) discusses spatial variability in soil denitrification rates and found that "hot-spots" of denitrification activity could be associated with particulate organic carbon material in the soil. Manure, being readily decomposable, enhanced denitrification (Rolston et al., 1979) while in another study wheat straw addition reduced denitrification and increased immobilization (Craswell, 1978). Plots receiving surface corn stover supplement had a 3 to 43 fold increase in denitrifying organisms (Doran, 1980b). Increasing the amount of organic mulch present on no-till or reduced till plots results in less evaporation and therefore increased soil moisture content. Koskinen and Keeney (1982) speculate that in carbon limiting systems, the rate of organic carbon mineralization controls denitrification to a greater extent

than pH. Dick (1983) has shown that no-tillage practices result in higher organic carbon and nitrogen concentrations at the soil surface. Carter and Rennie (1983) have demonstrated that net mineralizable carbon and nitrogen were greater in the surface of a zero-tillage system. Bremner and Shaw (1958a,b) observed that denitrification occurred in soils with <1% native soil carbon, yet the availability of this carbon and not just the total organic carbon determines the activity. Burford and Bremner (1975) determined that water soluble carbon could account for about 70% of the nitrous oxide and nitrogen evolved during soil denitrification. They concluded that quantification of mineralizable carbon or water soluble organic carbon was an indicator of soil denitrification capacity. deCatanzaro and Beauchamp (1985) agree that mineralizable carbon does allow prediction of denitrification rates but only for the early stages (the first four days) of the experiment. A study of the effects of carbon, moisture, and nitrate concentration on denitrification in a clay loam soil indicated that differences in C addition increased denitrification activity by 40 to 63% while moisture changes and nitrate concentration increase had minimal effects (Myrold and Tiedje, 1985). Incorporation of straw into conventionally tilled soil or a surface application on zero tilled soil plots with adequate inorganic N present approximately

doubled the denitrification losses over nonamended plots. This was apparently caused by the increased supply of residue and the increased surface moisture content especially in the zero tilled plots (Aulakh et al., 1984). Zero tilled soil plots had two times the nitrogen loss of the conventionally tilled soil plots as determined by acetylene inhibition. On plots with straw, nitrogen sources, or nitrogen plus straw added, the zero tillage plots had three times the nitrogen loss of the conventionally tilled plots. Oxygen presence in the profile decreased  $N_2O$  production by soil organisms (Firestone et al., 1979). Other authors indicate that denitrification increased following a decrease in air filled porosity after rainfall (Aulakh, 1982; Sexstone et al., 1985a) and they reported the greatest  $N_2O$  production immediately following irrigation.

#### H. TEMPERATURE AND pH

Temperature and pH also affect denitrification. Denitrifiers appear able to adapt to a wide range of soil temperatures. In several studies (Focht, 1974; Keeney et al., 1979; Freney et al., 1979) temperatures up to  $60^{\circ}C$  were used to incubate soils and denitrification was observed in all cases. The denitrification found at  $60^{\circ}C$  was attributed to the presence of preformed enzymes. Lower temperatures of incubation (10 to  $15^{\circ}C$ ) caused an abrupt decrease in denitrification and at  $0^{\circ}C$  denitrification became imperceptible

(Stanford et al., 1975b). Bonrud (1985) in a temperature study found that maximum denitrification rates occurred at 37°C. Craswell (1978) indicates that at lower temperatures (10°C or less), oxygen is more soluble in water and diffuses readily, therefore, the oxygen may inhibit denitrification when the soil reaches these lower temperatures. Focht (1974) states that two functions, aeration and pH, cause the most variability in  $N_2O$  production. Soils with pH's adjusted from 4 to 8 were used to demonstrate that denitrification potential increased with increasing pH (Focht, 1974; Keeney et al., 1979). No response to changes of pH from 5 to 6 could be seen by Davidson and Swank (1987). Muller et al. (1980) were able to show that even at low pH's, denitrification could be shown to occur on forested, miry, and agricultural soils in Finland. They determined that vegetation type had little influence on denitrification rate. A low pH (<4.5) enhanced the production of NO and  $N_2O$  gases and may have caused underestimation of denitrification rate. At a low pH (<4.8), Bremner and Shaw (1958) found denitrification proceeded very slowly and increased with a rise in pH to a very rapid rate at pH 8.0 to 8.6. Weier and Gilliam (1986) suggest that at low pH, the nitrous oxide reductase enzyme is inactive, therefore, losses of nitrogen are in the form of  $N_2O$ , while at neutral to high pH, the losses occur in the form of  $N_2$  gas. There is evidence which



suggests that a slightly alkaline pH favors maximum denitrification (Bremner and Shaw, 1958; Nommik, 1956).

Eastern South Dakota contains a wide variety of soil types and parent materials. This experiment was conducted as part of an integrated study sponsored by the USDA, Eastern South Dakota Area, and Water Research Team in Lake County (see Figure 1).

The distribution of soil mapping units was mapped for the mapping unit in the research area (N 1/4, SW 1/4, Sec. 35, T.142 N., R.51 W.) is shown in Figure 2. This experiment involved two soils, a Rendzina soil (Fertile, nonacidic, more typical Argosol) and a Mollisol soil (Fertile, nonacidic, more typical Argosol, more typical). (Soil Survey Staff, 1973). These soils are typical of those found in the northern portion of this Eastern South Dakota area.

The Rendzina soil is a very light brown, silty clay loam, with a high water content and a high organic content. It is a very fertile soil, with a high water content and a high organic content. The Mollisol soil is a very light brown, silty clay loam, with a high water content and a high organic content. It is a very fertile soil, with a high water content and a high organic content. The Rendzina soil is a very light brown, silty clay loam, with a high water content and a high organic content. It is a very fertile soil, with a high water content and a high organic content. The Mollisol soil is a very light brown, silty clay loam, with a high water content and a high organic content. It is a very fertile soil, with a high water content and a high organic content.

## MATERIALS AND METHODS

### I. EXPERIMENTAL PLOTS

Eastern South Dakota contains a wide variety of soil types and parent materials. This experiment was conducted as part of an integrated project occurring at the USDA Eastern South Dakota Soil and Water Research Farm in Lake County (see Figure 1).

The distribution of soil mapping units and legend for the mapping units at the research farm (E 1/2, NW 1/4, Sec. 35, T.107 N., R.53 W.) is shown in Figure 2. This experiment involved two soils, a Beadle silt loam (Fine, montmorillonitic, mesic Typic Argiustoll) and a Worthing silty clay loam (Fine, montmorillonitic, mesic Typic Argiaquoll) (Soil Survey Staff, 1973). These soils are typical of those farmed on the morainic landscapes of this Late Wisconsin age deposited material.

The Beadle soil is a deep well drained nearly level to undulating upland loamy soil with a clayey subsoil. It was formed in glacial till on the uplands. The Worthing soil is typically found in drainageways or flat, enclosed depressions. These alluvial soils are poorly drained and level. They are silty soils with clayey subsoils. There is poorer drainage than that found on the Beadle soil, and indeed, in 1986, this soil was extremely wet most of the year.

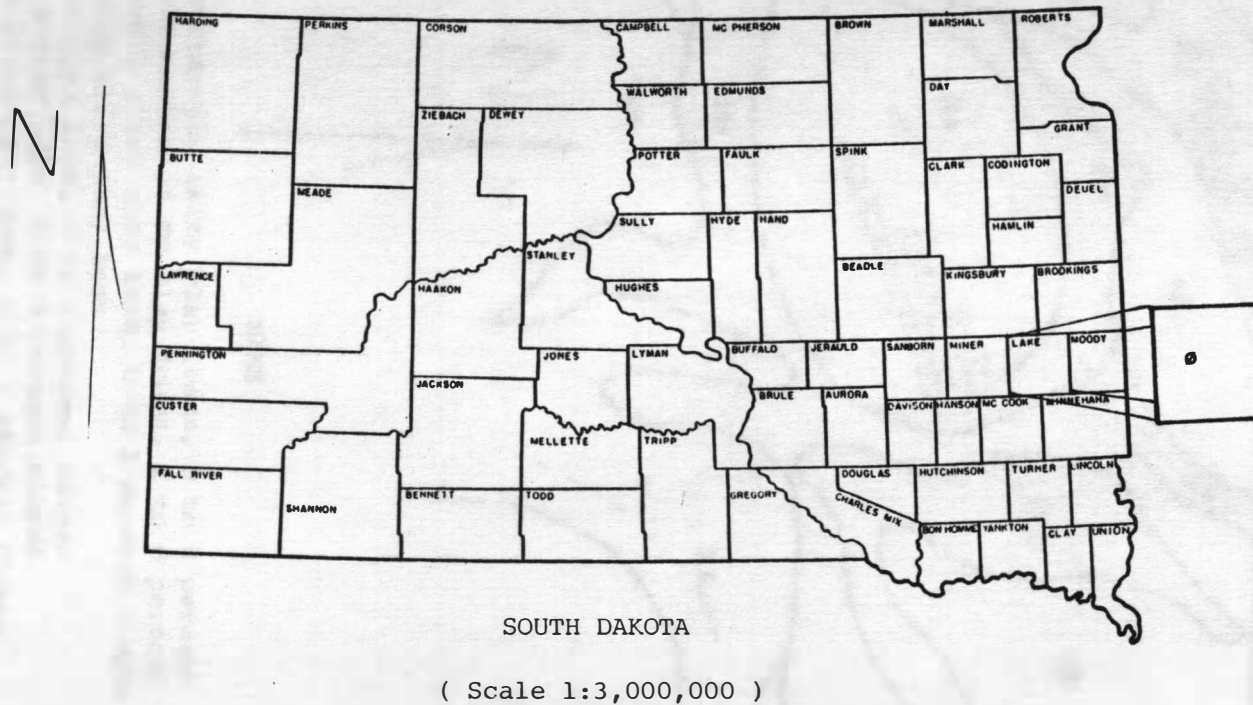
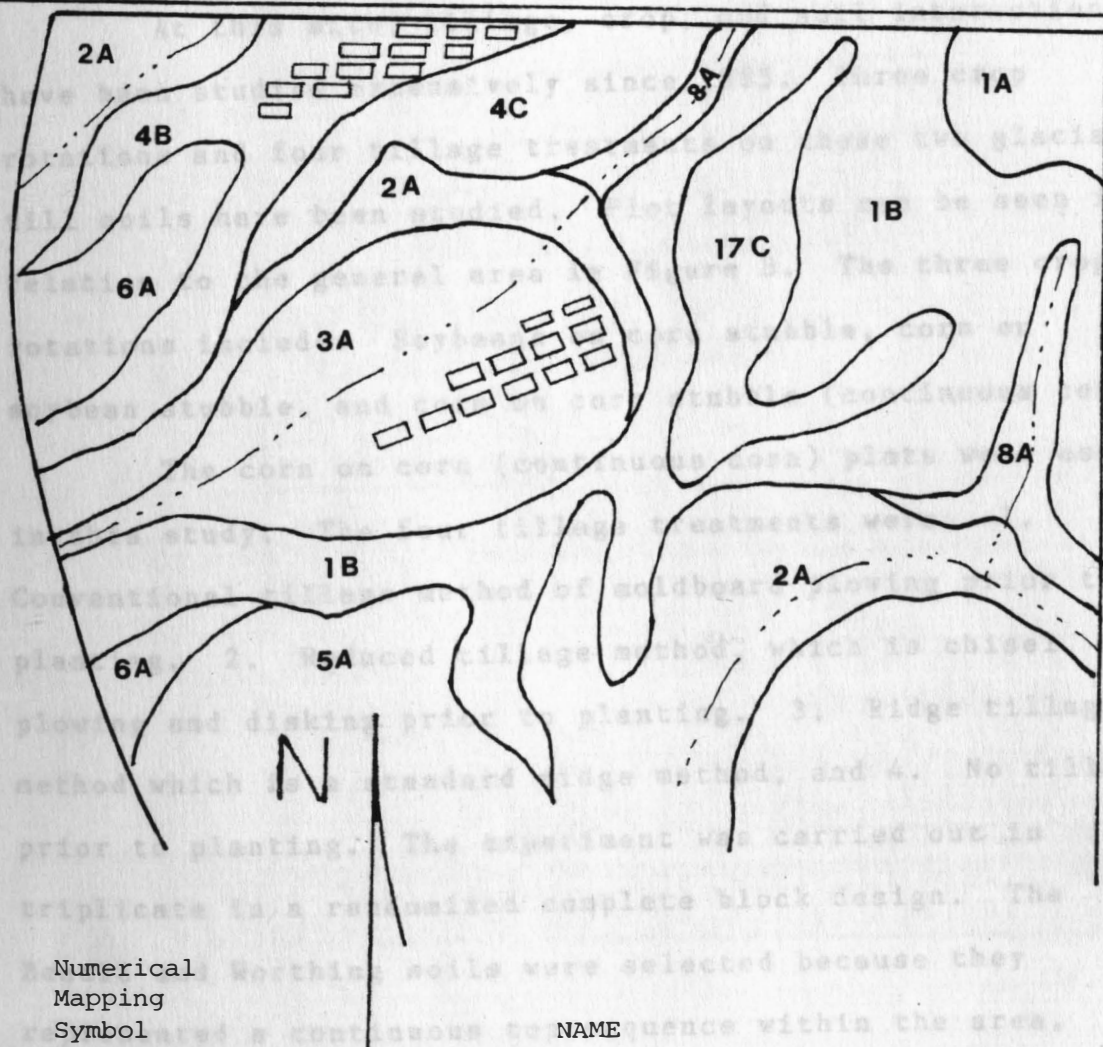


Figure 1. Location of the USDA Eastern South Dakota Soil and Water Research Farm in Lake County, South Dakota.



- 1A Wentworth-Egan silty clay loams, 0 to 2 percent slopes
- 1B Egan-Wentworth silty clay loams, 2 to 6 percent slopes
- 2A Whitewood silty clay loam, 0 to 2 percent slopes
- 3A Worthing silty clay loam
- 4B Beadle clay loam, 2 to 6 percent slopes
- 4C Beadle clay loam, 6 to 9 percent slopes
- 5A Sinai silty clay loam, 0 to 2 percent slopes
- 6A Wentworth silty clay loam, 0 to 2 percent slopes
- 8A Viborg silty clay loam, 0 to 3 percent slopes
- 17C Egan-Beadle complex, 6 to 9 percent slopes

Figure 2. Soils on the research farm, plot layout, and map unit descriptions.



At this site, tillage, crop, and soil interactions have been studied extensively since 1985. Three crop rotations and four tillage treatments on these two glacial till soils have been studied. Plot layouts can be seen in relation to the general area in Figure 3. The three crop rotations include: Soybeans on corn stubble, corn on soybean stubble, and corn on corn stubble (continuous corn).

The corn on corn (continuous corn) plots were used in this study. The four tillage treatments were: 1. Conventional tillage method of moldboard plowing prior to planting. 2. Reduced tillage method, which is chisel plowing and disking prior to planting. 3. Ridge tillage method which is a standard ridge method, and 4. No tillage prior to planting. The experiment was carried out in triplicate in a randomized complete block design. The Beadle and Worthing soils were selected because they represented a continuous toposequence within the area, ranging from the upland and shoulder Beadle soil to the toeslope Worthing soil. This represents a typical farm landscape in eastern South Dakota. When farmed, the soils are not tilled independently of each other, but are farmed as a unit. Therefore, both soils are included in this study. Pioneer 3747 MF seedcorn was planted at 26,000 plants per acre. Fertilizer (12-36-12) was applied at 104 pounds per acre. Ten pounds of Carbofuran insecticide and

## Beadle Soil - Tillage Experiment

II-1			I-1			III-1		
SB	**	C	SB	**	C	SB	C	**
9	8	7	6	5	4	3	2	1
II-2			I-2			IV-1		
C	SB**		SB	C	**	C	SB	**
18	17	16	15	14	13	12	11	10
III-3			IV-2			III-2		
**	SB	C	**	SB	C	C	SB	**
27	26	25	24	23	22	21	20	19
II-3			I-3					
**	C	SB	**	SB	C			
33	32	31	30	29	28			
IV-3								
**	SB	C						
36	35	34						

## Worthing Soil - Tillage Experiment

			III-3			IV-3		
			C	SB	**	SB	C	**
			36	35	34	33	32	31
			II-2			I-3		
			C	SB	**	**	C	SB
			30	29	28	27	26	25
			II-2			I-1		
			C	SB	**	C	SB	**
			24	23	22	21	20	19
III-2			IV-2			III-1		
SB	C	**	C	**	SB	SB	C	**
18	17	16	15	14	13	3	2	1
			IV-1					
			**	C	SB			
			12	11	10			
			II-1					
			C	**	SB			
			9	8	7			
			I-1					
			C	SB	**			
			6	5	4			

CROP

C = Corn (on previous soybean plot - 1985)  
 SB = Soybean (on previous corn plot - 1985)  
 \*\* = Continuous Corn - Denitrification samples taken in these plots

TREATMENTS

I = Conventional Tillage	-1 = Rep 1
II = Reduced Tillage	-2 = Rep 2
III = Ridge Tillage	-3 = Rep 3
IV = No Tillage	

Figure 3. Cooperative plots

ten pounds of Propachlor herbicide were applied at planting. Alachlor (2.5 quarts per acre) and Cyanazine (2 quarts per acre) were sprayed onto the plots in midseason.

Soil moisture at planting and bulk density were determined by standard methods (Singer and Munns, 1987, Allison, 1965). Residue measurements were made immediately after planting by counting the number of residue points on four 25 point grids per plot (Hartwig and Loflen, 1978). Soil temperature at 10 cm (planting depth) was available for the growing season. Yield measurements were also taken at the end of each growing season (B.R. Khakural, Ph.D. Dissertation, in progress).

The Beadle soil corn plots were planted on May 20, 1986. There was sufficient moisture in the seedbed for germination and the rainfall pattern the rest of the summer was such that there were few periods of insufficient moisture for good crop growth. Contrary to this, on the Worthing soil, planted May 21, 1986, the conventionally tilled plots were extremely wet. While the other plots could be planted, these could not. Planting of two of the three replications of these plots was carried out with a smaller planter on a later date. The third replication could not be planted, and indeed, the water ponded on this plot all summer. There were very few days on which the Worthing soil sites were not muddy in 1986, since during

April through September 81.66 cm of rain were recorded.

In 1987, the planting date was May 5 for the Beadle soil and May 6 for the Worthing soil. The 1987 moisture pattern followed a more typical trend for this portion of South Dakota, with the area receiving 37.72 cm of precipitation from April through September. Thus, 1986 was a wet year and 1987 was a normal year from the precipitation perspective.

## II. SOIL SAMPLE COLLECTION

Soil samples were collected only from the continuous corn rotation plots for analysis of denitrification ability. These samples were collected biweekly from April until October for the 1986 and 1987 growing season. Soil samples were collected by means of a 2.5 x 5 cm-soil probe as described by Bonrud (1985). Bonrud determined that a 0- to 5-cm depth soil sample would allow accurate determination of a denitrification rate. At sampling, two soil cores, one from within the corn row (between two corn plants) and one from between the rows, were placed into each 50-cc plastic syringe. One syringe was prepared from each plot. The syringe was stoppered with a serum stopper at the tip and a #6 rubber stopper where the plunger is normally positioned. Each syringe was labelled with the soil, tillage, and replication before being placed into a cooler filled with ice to retard microbial activity. After all the samples



were collected, the cooler containing them was taken back to the microbiology laboratory at SDSU, where the denitrification analysis was performed.

### III. SAMPLE PREPARATION

In the microbiology laboratory, each syringe was connected to a gassing manifold and the air in the syringe was evacuated to 0.9 atm and replaced with argon gas. The syringes were each evacuated and refilled three times. A 3.5 ml portion of the argon atmosphere was removed and replaced by 3.5-ml acetylene to inhibit denitrification of nitrate beyond nitrous oxide. Bonrud (1985) shows that using the gassing manifold in this manner would optimize denitrification and that the 3.5 ml of acetylene would provide a five percent acetylene atmosphere, which best inhibited the conversion of nitrous oxide to nitrogen gas. The 1986 samples were incubated in a closed container for 24 hours (for temperatures, see Appendix E) while the 1987 samples were incubated at the average monthly temperature for Lake County for the month in which they were collected (See Appendix E). The monthly high and monthly low temperatures were used to determine the average monthly temperature for incubation. Bonrud (1985) demonstrated in a laboratory study that soil denitrification rate increased at temperatures over 8 °C up to 60 °C.

#### IV. GAS CHROMATOGRAPHY

After the 24-hour incubation, an aliquot of the airspace in the syringe (up to 0.25 ml) was injected into a Varian 3700 gas chromatograph equipped with a  $^{63}\text{Ni}$  source in an electron capture detector. The gas chromatograph injection port temperature was  $100^{\circ}\text{C}$ , the column temperature was  $75^{\circ}\text{C}$ , and the detector temperature was  $320^{\circ}\text{C}$ . A gas flow rate of  $40\text{ ml min}^{-1}$  was maintained. The 2-m x 0.3-cm stainless steel column within the chromatograph was filled with 50/80 Poropak Q packing material (Supelco Inc., Bellfonte, Penn.). A Spectra Physics 4270 integrator was programmed to determine  $\mu\text{L L}^{-1}$  nitrous oxide proportional to a standard nitrous oxide injection.

A 30- $\mu\text{L}$  sample ( $29.85\text{ }\mu\text{L L}^{-1}$ ) nitrous oxide was injected as the nitrous oxide standard (Dakota Welding, Watertown, South Dakota). Integrator response to the nitrous oxide standard was used to calculate the nitrous oxide concentration in unknown samples. After every six injections, the gas standard was used to recalibrate the integrator. A gas sample (up to 0.25ml) from the airspace of each of the syringes was injected into the gas chromatograph and the integrator determined the  $\mu\text{L L}^{-1}$  nitrous oxide in the amount injected. If the nitrous oxide exceeded  $30\text{ }\mu\text{L L}^{-1}$  in any unknown sample, a smaller aliquot of that same sample was injected so that a value within the

20- to 30-uL L<sup>-1</sup> range could be reported. If the nitrous oxide did not approach this range upon an injection of up to 0.25 ml of sample, the value obtained for the 0.25-ml injection was recorded as the result of that samples analysis. There appeared to be a limit of 0.25 ml which could be injected, due to the moisture and other contaminants present in the air samples which produced interfering peaks when present in large quantities.

#### V. SOIL MOISTURE CONTENT AND pH

The amount of water in each soil sample was determined for use in the calculation of percent volumetric water and in the calculation of the amount of water in the syringe, which absorbed nitrous oxide. The amount of water in each sample was determined by a weight loss method. The two stoppers were removed from each syringe and the soil plus syringe weight was recorded. The weighed syringe-soil samples were placed into a Frigidaire model MC 700 M microwave oven. The samples were dried at power level six for one hour (or longer) with frequent turning to prevent melting of the syringes (one turn every three to four minutes) until moisture could no longer be seen on the inside of the syringe or in the tip of the syringe (Gee and Dodson, 1981). The syringe-soil samples were placed into a dessicator to cool prior to weighing them to determine the moisture loss. The soil sample weight was the total weight



minus the the average empty syringe weight (12.77 grams).

A 1:1 soil:water solution, allowed to equilibrate for 20 minutes was used to determine soil pH (Peech et al., 1947).

VI. CALCULATION OF DENITRIFICATION (kg nitrogen per hectare per day).

The amount of nitrogen lost by denitrification was calculated for each sample. A Supercalc3 spreadsheet program (Sorcim) was used for data handling and manipulation. Data entries included syringe-sample weights before and after microwaving,  $\mu\text{L L}^{-1}$  nitrous oxide from the integrator printout, and an injection factor (which is  $1/\text{amount injected}$ , to transform the data to a per ml of gas injected basis). The program corrected for bulk density of the soil samples (Blake, 1965) and percent volumetric moisture. The bulk density values obtained were an average of the bulk density in the row and between the row, since both cores were combined in one syringe. Nitrogen losses were converted to kg nitrogen loss per hectare per day by the calculations shown below:

- A. Standard values used in the calculations below are:
- average syringe weight = 12.77 g
  - total volume of syringe = 70 ml
  - volume of two cores =  $50.64 \text{ cm}^3$
  - particle density =  $2.65 \text{ Mg m}^{-3}$  (Donahue et al.,

1983)

amount of dissolved  $N_2O$  per ml water = 0.676

Number of cores per syringe = 2

Ground surface area of one core ( $m^2$ ) =  $4.52 * 10^{-4}$

ml of gas per liter = 24560

Molar amount of N in  $N_2O$  = 0.028

- B. Bulk density, percent moisture by weight, percent moisture by volume and porosity were determined for each soil sample by standard methods as shown below.
- bulk density ( $Mg\ m^{-3}$ ) = (dry weight of soil plus syringe minus the average syringe weight) divided by the volume of the two cores.
  - Percent water by weight = [(wet weight - dry weight of soil) times 100] divided by the dry weight of the soil.
  - Percent water by volume = (percent water by weight)\*(bulk density)
  - porosity =  $100 - (\text{bulk density}/\text{particle density})$  times 100)
- C. Air filled volume and air filled pores are calculated in order to determine ml air per syringe as shown below:
- Percent air filled volume = porosity - percent water by volume



b. Air filled pores in the soil = volume of the two cores minus percent air filled volume divided by 100)

c. ml air per syringe = total volume of syringe minus total volume of the two cores plus air filled pores in the soil.

D. The nitrous oxide emissions were reported by the integrator in  $\mu\text{L L}^{-1} \text{N}_2\text{O}$  ( $\mu\text{L L}^{-1} \text{N}_2\text{O}$  injection). This was converted to ml  $\text{N}_2\text{O}$  per sample and ml  $\text{N}_2\text{O}$  per day. The amount of dissolved nitrous oxide and other manipulations of the data are shown below in order to arrive at a value of  $\text{kg N}_2 \text{ha}^{-1} \text{day}^{-1}$  emitted from the soil.

a. ml  $\text{N}_2\text{O}$  per sample = recorded injection  $\mu\text{L L}^{-1}$  divided by  $10^6$

b. ml  $\text{N}_2\text{O}$  per sample per day = (ml air per syringe) \* (ml  $\text{N}_2\text{O}$  per sample) divided by mls injected

c. ml  $\text{N}_2\text{O}$  per sample per day plus dissolved  $\text{N}_2\text{O}$  = (amount of dissolved  $\text{N}_2\text{O}$  per ml water) times (wet minus dry wt of soil) divided by (ml air per syringe plus ml  $\text{N}_2\text{O}$  per sample per day)

d. ml  $\text{N}_2\text{O}$  per ha per day = (ml  $\text{N}_2\text{O}$  per sample per day plus dissolved  $\text{N}_2\text{O}$ ) divided by (the number of cores per syringe) divided by (the ground surface area of one core)

e. moles  $N_2O$  per ha per day = (ml  $N_2O$  per ha per day times  $10000m^2$  per ha) divided by ml gas per liter

f.  $kg N_2 ha^{-1} day^{-1}$  = moles  $N_2O$  per ha per day times the molar amount of N in  $N_2O$

## VII. MOISTURE CONTROL BY TENSION TABLE

### A. Effect of aggregate size on denitrification

Soil samples from the top 5 cm of each plot on the Worthing soil were collected, air dried and sieved into five particle sizes (0 to 1 mm, 1 to 2 mm, 2 to 4.75 mm, 4.75 to 6.2 mm, and 6.2 to 9 mm). The soil was placed in the top of a nest of sieves and gently shaken until no soil passed through the smaller sieve. Two one-inch rings (one-inch sections of syringe barrels) were placed on an Eijkelkamp sand table (tension table) and filled with soil of one aggregate size. The tension table is equipped with a hanging water column which allows adjustment of tension on samples placed on the table. The table was filled with samples, each two rings containing a different size aggregate or soil from a different plot. The samples were saturated with water and the hanging water column adjusted to 50-cm depth. Fifty cm was chosen because the resulting soil moisture was near that found under field conditions. Moisture in the samples was allowed to equilibrate for three days on the tension table before the soil in each pair of

rings was removed and placed into a syringe, labelled, and stoppered. The syringes were taken to the microbiology laboratory, evacuated, and acetylene added. The syringes were incubated at 17°C prior to analysis by gas prepared chromatography. Of these four syringes, one was opened and

#### B. Equilibration on the tension table while containing

Samples of soil from replication three of the Worthing ridge tilled soil (unsieved) were placed in the one inch rings on the sand table, saturated, and the tension was adjusted to 50 cm. Four rings were removed at regular time intervals. Two rings of soil were placed into each syringe, labelled, and gassed for gas chromatographic analysis. the Samples were removed at regular intervals between 6 and 78 hours after initiation of the experiment. Samples were placed two per syringe, gassed with argon, acetylene added as before, and incubated 24 hours at 20°C. The analysis was done in duplicate.

#### C. Stimulation of denitrification by glucose and nitrate.

A sample of soil from the top 5 cm of replication three of the ridge till plots on the Worthing soil was dried and sieved. Samples of twelve grams each from the 2- to 4.75-mm fraction were placed in rings on a sand table. Five ml of an aqueous solution of glucose, nitrate, glucose plus nitrate, or control (water alone) were added to each soil sample to saturate it. The tension table was then adjusted



to 50-cm tension for 24 hours. Two rings of soil were then placed into a 50-cc syringe. Eight soil samples of each control or glucose and/or nitrate treatment had been placed on the sand table, therefore, four syringes were prepared from each set. Of these four syringes, one was opened and refrigerated to allow the soil to dry while curtailing bacterial activity. The remaining three syringes were gassed with argon, 3.5 ml replaced with acetylene, and incubated at 20 °C for 24 hours prior to gas chromatographic analysis for nitrous oxide production.

This experiment was performed in three parts. First, only nitrate solutions were added to the soils, the second time, only glucose solutions were added, and the third time, mixtures of glucose and nitrate were added. Each experiment was repeated once.

#### VIII. GLUCOSE ASSAY ON PRE- AND POST- CULTIVATION SOIL SAMPLES .

A glucose equivalent assay, using an anthrone method as described by Stanford et al. (1975a), was performed on 5-g soil samples. One set of soil samples had been collected from the top 5 cm of soil in each rep of each plot prior to cultivation for weeds (July 8, 1987). An equal amount of soil was collected from in the rows and between the rows of corn. Soil samples were collected from the no tillage plots at the same time. Equal amounts of soil were

collected from in the row and between the row. Post-cultivation samples were collected on August 10, 1987. These samples were air dried and nitrate analysis was performed on them by the SDSU Soil Testing Lab (Carson, 1975). A second set of samples was collected in early September, after tillage. In each case, these samples were air dried and crushed. Duplicate five-gram samples were placed into two 50-cc plastic tubes, 25 ml of 0.01M  $\text{CaCl}_2$  was added to each, the tubes were fitted with rubber stoppers (#6) with 30-cm glass tubes (condenser) through them. The entire system (tub, stopper, and glass tube) was placed in a steamer and boiled for one hour. The resulting soil suspension was cooled and centrifuged in a tabletop centrifuge (International Clinical Centrifuge model CL, rotor catalog number 809) for fifteen minutes at setting 6. One ml of the supernatant fluid was removed to a clean test tube for glucose equivalent analysis (Stanford et al., 1975a).

At the same time as the samples are being prepared, a set of standard glucose samples are prepared. Glucose was added to 0.01M  $\text{CaCl}_2$  to make a 100-ug-per-ml glucose stock solution. Standards of 0, 12.5, 25, 50, 75, and 100 ug per ml are made in test tubes by dilution of the stock solution. The volume in each test tube is adjusted to one ml to keep all measurements equal. Water is used as a blank in the



spectrophotometer cuvette. The set of standards was prepared for each time the analysis was performed. A two percent solution of anthrone (Sigma) in ethyl acetate was freshly prepared and 0.5 ml added to the standards and unknown supernatants. The prepared anthrone reagent could be stored for 5 to 7 days in the dark. Five ml of concentrated sulfuric acid was layered over the mixture. The tube was swirled gently so that violent boiling upon mixing of the acid and water layers was avoided. The tube was then mixed thoroughly. Each tube was allowed to stand at room temperature for 10 to 15 minutes before the percent transmittance at 620-nm wavelength was recorded from a Bausch and Lomb spectrophotometer model 340. The spectrophotometer reading was compared to the data obtained from the standard glucose curve described above. Glucose in  $\mu\text{g ml}^{-1}$  was determined from the linear regression,  $\mu\text{g glucose g}^{-1}$  soil could then be calculated. The reading for the 0- $\mu\text{g}$  glucose standard was subtracted from all readings prior to analysis. In all cases, the  $r^2$  for the linear regression of the spectrophotometer reading vs. amount of glucose was 0.985 to 0.999.

#### IX. ANALYSIS OF VARIANCE

The analysis of variance results can be seen in Appendix F. Data analyses of denitrification rate, volumetric moisture, and bulk density were performed using

tillage, soil, replication, and, when appropriate, year as class variables. The Proc GLM (General Linear Models) of SAS (Statistical Analysis Systems) was used for analysis of variance when missing cells were involved in the analysis, otherwise, Proc ANOVA (Analysis of Variance) was conducted in a randomized complete block design at two locations (soils). The class variables used in the analyses consisted of soil (Beadle or Worthing), four tillage treatments (conventional tillage, reduced tillage, ridge tillage, and no tillage) and three replications. Each value reported for a replication is an average of three observations. The dependent variable is denitrification rate ( $\text{kg N}_2 \text{ ha}^{-1} \text{ day}^{-1}$ ).

The analysis of aggregate size denitrification involved tillage and replication as the class variables and was conducted in a randomized complete block design on five aggregate sizes (0 to 1 mm, 1 to 2 mm, 2 to 4.75 mm, 4.75 to 6.36 mm, and 6.36 to 9.53 mm). A multivariate analysis of variance was performed by aggregate size fraction. The class variables were one soil (Worthing), four tillage treatments (conventional tillage, reduced tillage, ridge tillage, and no tillage). The dependent variable is denitrification rate.

The analysis of variance for the glucose and nitrate content of the soil pre- and post-cultivation was performed

by using soil (Beadle or Worthing), tillage (conventional tillage, reduced tillage, ridge tillage, and no tillage), replication and sample time (pre- or post- weed cultivation) as the class variables. The dependent variable was glucose or nitrate content.

In the analysis of variance for the dependent variables residue, bulk density, volumetric moisture, or denitrification rate, the class variables were soil (Beadle or Worthing), tillage (conventional tillage, reduced tillage, ridge tillage, and no tillage), and year (1986 and 1987).

Soil samples were collected from the ridge tillage plots on July 14, 1986 (Figure 1). In 1987 (Figure 2), high peaks of 2- to 4-kg nitrogen application occurred on the ridge and conventional tillage plots throughout the growing season on the Beadle soil.

On the Worthing soil, nitrogen was not applied. Denitrification varied between 0 and 1.5-kg nitrogen applied (Figure 3). The highest levels of denitrification occurred on the ridge tillage plots in 1986 and 1987. In 1986, the highest level of denitrification was 1.5-kg nitrogen applied on the ridge tillage plots. There was no denitrification on the conventional tillage plots. High values of 3- to 4-kg nitrogen applied were observed on the ridge tillage plots in 1987. A peak of 1.5-kg nitrogen applied occurred in the ridge tillage plots on July 14, 1986 for the Worthing soil. The Worthing soil showed denitrification levels of 2- to

## RESULTS AND DISCUSSION

### I. FIELD STUDIES

#### A. Denitrification Rate by Tillage

Denitrification rates in each of the two soils studied exhibited wide variances. The average denitrification occurring with each tillage method on each sampling date throughout 1986 and 1987 for the Beadle soil varied between 0- and 5-kg nitrogen evolved  $\text{ha}^{-1} \text{ day}^{-1}$ . While on some days some plots did not show any denitrification losses, a high of nearly 5-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$  evolved from the ridge tillage plots on July 15, 1986 (Figure 4). In 1987 (Figure 5), high peaks of 2- to 4-kg nitrogen evolution occurred on the ridge and reduced tillage plots throughout the growing season on the Beadle soil.

On the Worthing soil, during the two years, denitrification varied between 0- and 5.6-kg nitrogen evolved (Figure 6). The highest amount of denitrification on any one date in 1986 on the Worthing soil, as on the Beadle soil, occurred on the ridge tillage plots. There was no definite pattern among these data. High values of 3- to 4-kg nitrogen evolved per hectare per day appear in the reduced tillage plots for both soils on June 2, 1986. A peak of 5.6-kg nitrogen evolved  $\text{ha}^{-1} \text{ day}^{-1}$  occurred in the ridge tillage plots on July 1, 1986 for the Worthing soil. The Worthing soil produced denitrification levels of 2- to

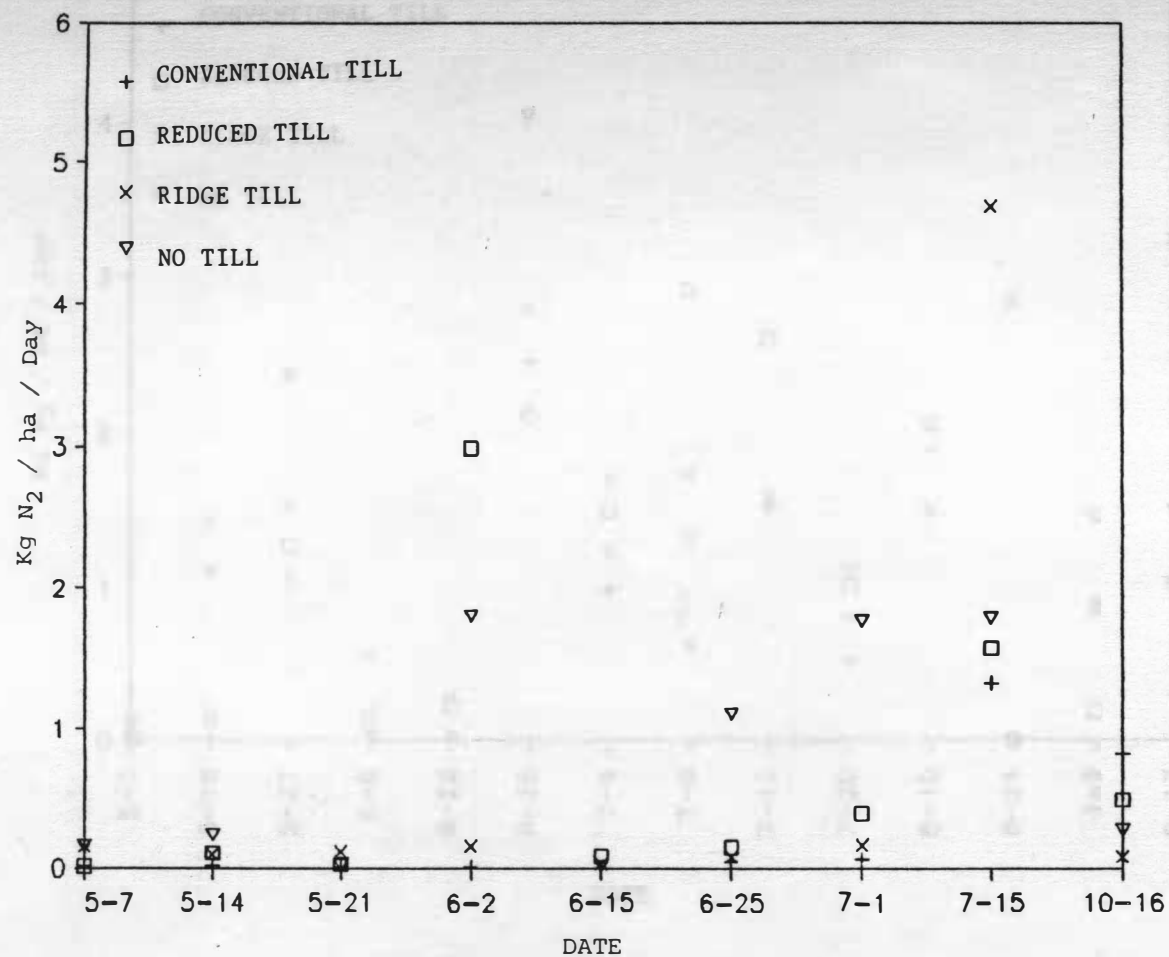


Figure 4. Denitrification on the Beadle Soil, 1986. Each plotted point is an average of three observations



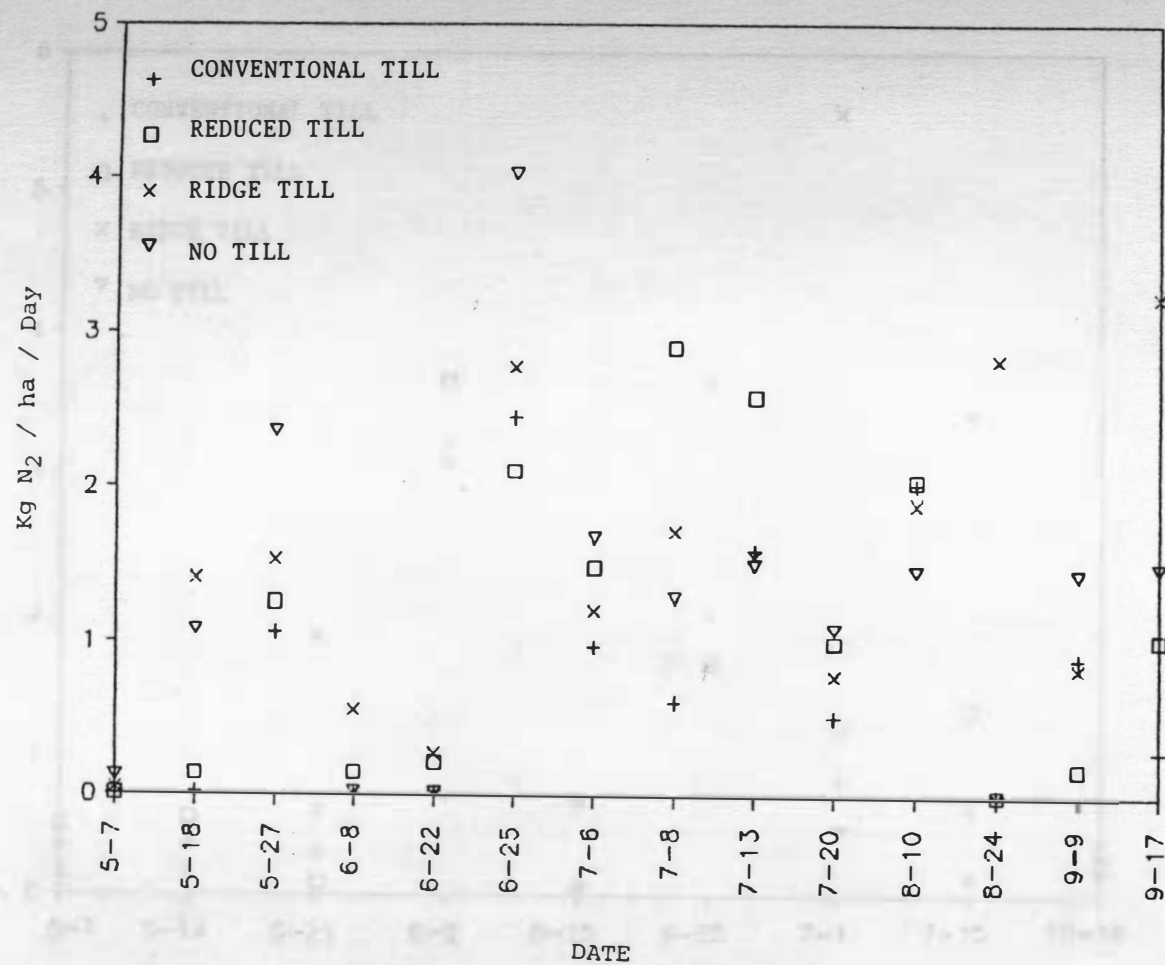


Figure 5. Denitrification on the Beadle Soil, 1987. Each plotted point is an average of three observations

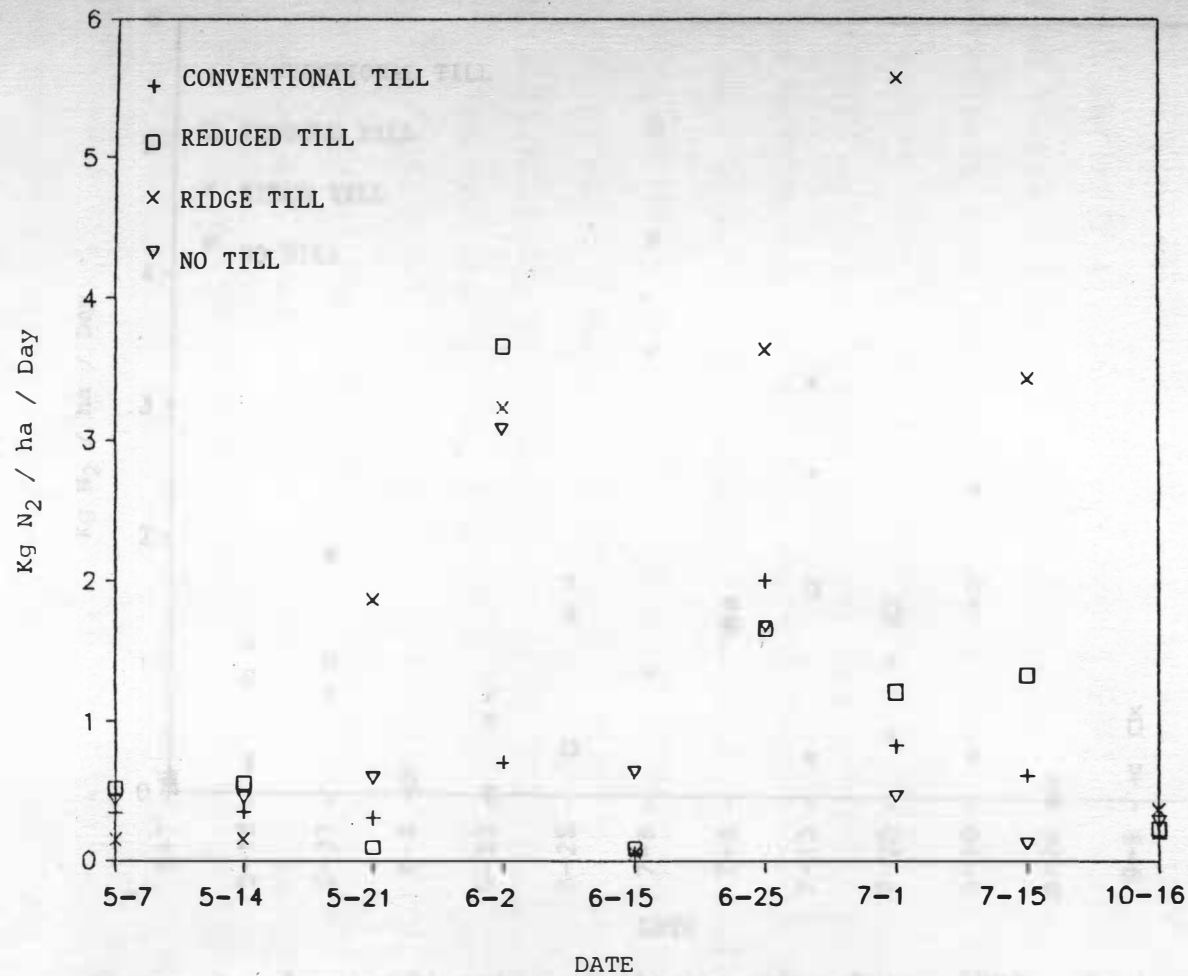


Figure 6. Denitrification on the Worthing Soil, 1986. Each plotted point is an average of three observations

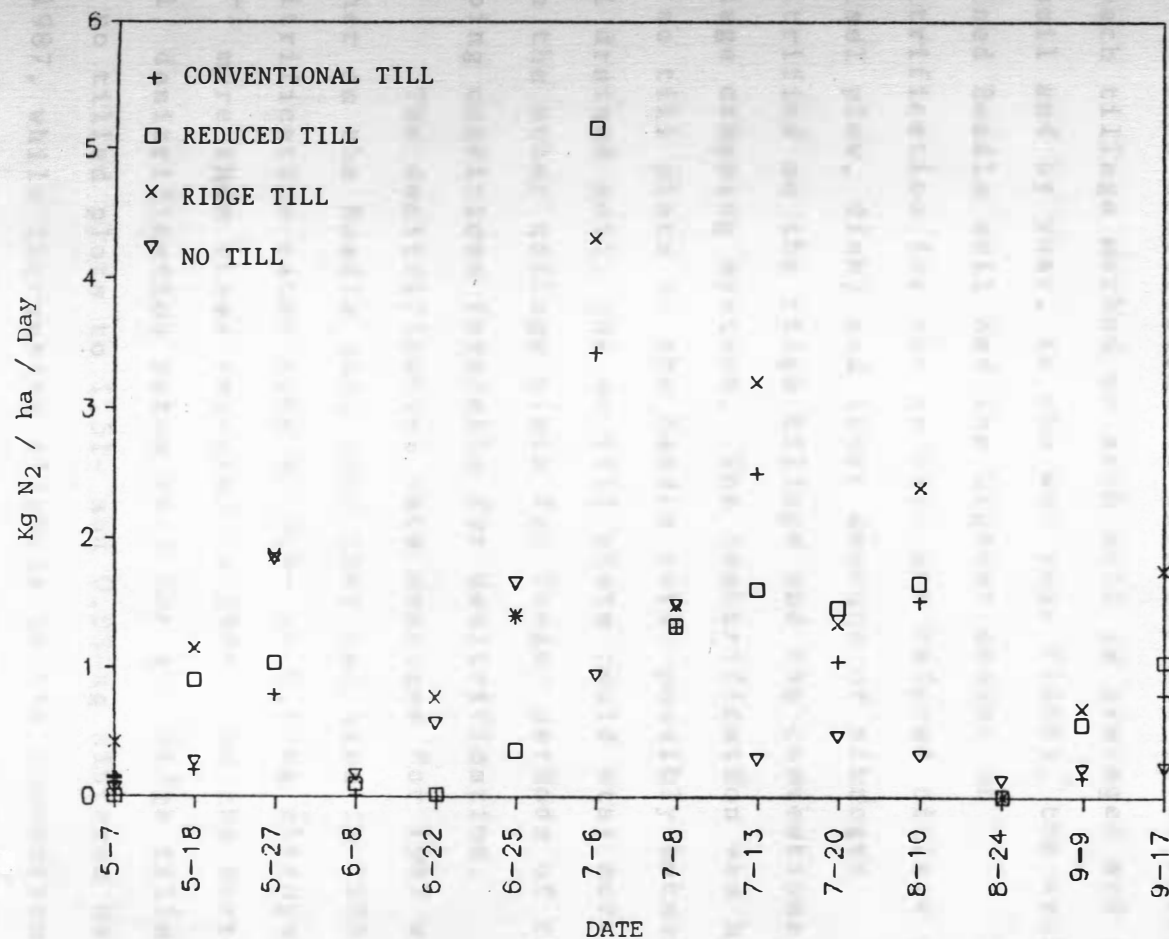


Figure 7. Denitrification on the Worthing Soil, 1987. Each plotted point is an average of three observations

5.2-kg nitrogen gas evolved  $\text{ha}^{-1} \text{ day}^{-1}$  for the 1987 growing season (Figure 7).

When denitrification data for the three replications of each tillage method on each soil is averaged and reported by soil and by year, in the wet year (1986), the well drained Beadle soil had the highest amount of denitrification for the no till and reduced tillage plots (chisel plow, disk) and lower amounts of nitrogen denitrified on the ridge tillage and the conventional tillage cropping systems. The denitrification was higher in the no till plots in the Beadle soil, possibly because on a well drained soil, the no till plots could stay more moist than the other tillage plots for longer periods of time, keeping conditions favorable for denitrification.

The denitrification rate averages for 1987 were higher on the Beadle soil than they had been in 1986. The denitrification rates rose by 0.4- to 0.8-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$  more than those reported in 1986. On the Worthing soil, denitrification rates fell for the ridge tilled plots and no tilled plots to 1.51- and 0.63-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$  in 1987, while increasing slightly in the conventionally tilled and reduced tilled plots to 0.96- and 1.12-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$ . The no and ridge tillage plots probably dried more in 1987 than in 1986. The denitrification rate decreased to a greater extent than that

in the conventional and reduced tillage plots, which dried in both years, since decreased residue cover on the conventionally tilled soil and soil stirring due to tillage, by both tillage methods, promoted drier conditions.

Denitrification in the Worthing soil in 1986 was large on the ridge tillage plots (averaging over 2-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$  for the sampling period) and lower amounts were observed on the conventionally tilled, reduced tilled, and the no tillage managed soils. On the poorly drained soil, conditions for denitrification are probably better on the ridge till systems due to the ridge protruding up out of the flooded areas, acting like a wick, which is not completely saturated and could pull moisture up out of the residue covered area between the rows, yet could warm up faster than the saturated conventional, reduced, or no till plots.

The wide variation in the data seen for denitrification on each of these soils might be due to a number of factors. Precipitation events greatly affect the denitrification. The increase of moisture decreases the air filled pore space and therefore aerobic organisms present in the soil that have less oxygen present. These organisms use up the oxygen rapidly, creating anaerobic pores allowing denitrification to take place.

The pH values of the Beadle soil varied considerably



more than those in the Worthing soil (Table 1). The pH of the Beadle soil no till plots decreased by 0.5 pH unit from 1986 to 1987. The ridge and conventional till plots pH decreased by 0.4 pH unit and the reduced till plots by decreased by 0.3 pH unit. On the Worthing soil, very small pH differences are found between the soil pH data obtained in 1986 and 1987. No difference is seen between the pH values of 1986 and 1987 for the reduced tillage Worthing plots. The pH of the soil rose 0.1 pH unit in the ridge tillage plots and fell by 0.1 pH unit in the no till plots. A decrease of 0.2 pH units was seen in the conventionally tilled Worthing plots. The pH of these soils was in the neutral to alkaline range and had no effect on the denitrification rate.

## B. Soil Physical Properties

### 1. Soil Moisture

Soil moisture has a large effect on denitrification. The denitrification rates in all tillage plots fell on July 20, 1987 to less than 1.6-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$ . A decrease of denitrification to near 0-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$  occurred on August 24, 1987 in all plots but one (denitrification in the ridge tillage plots in the Beadle soil increased to 2.5-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$ ). Both of these dates correspond to periods of low soil moisture, as can be seen in Figures 9 and 11. On June 2, 1986 (Figures 8

Table 1. pH averages 1986 and 1987 by soil and tillage

<u>Tillage</u>	<u>Beadle soil</u>		<u>Worthing soil</u>	
	<u>1986</u>	<u>1987</u>	<u>1986</u>	<u>1987</u>
Conventional	7.2	6.8	7.7	7.5
Reduced	7.8	7.5	7.6	7.6
Ridge	6.7	6.3	7.5	7.6
No till	7.0	6.5	7.7	7.6

No significant differences were detected.

Figure 8. Volucella data for 1986 and 1987. Each plant was observed.

and 10), the soils did not have an increase in soil volumetric moisture but the daily temperature had been increasing. This increase in temperature may have caused the increase in denitrification. Again, on July 15, 1986, no increase in moisture was seen (Figures 8 and 10), yet an

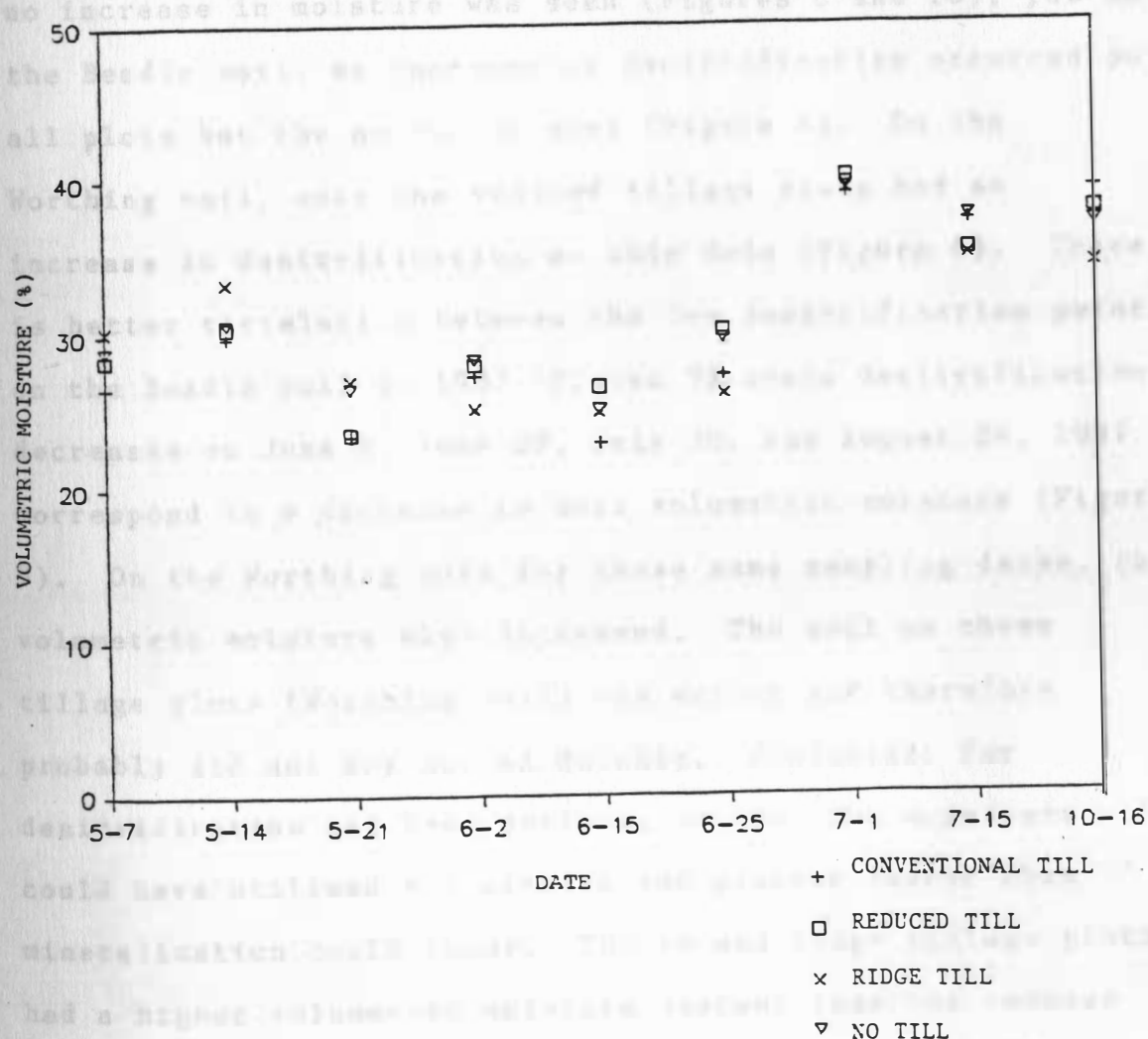


Figure 8. Volumetric moisture on the Beadle Soil, 1986.  
Each plotted point is an average of three observations.

and 10), the soils did not have an increase in soil volumetric moisture but the daily temperature had been increasing. This increase in temperature may have caused the increase in denitrification. Again, on July 15, 1986, no increase in moisture was seen (Figures 8 and 10), yet on the Beadle soil, an increase in denitrification occurred on all plots but the no tilled ones (Figure 4). On the Worthing soil, only the reduced tillage plots had an increase in denitrification on this date (Figure 6). There is better correlation between the low denitrification points on the Beadle soil in 1987 (Figure 5) where denitrification decreases on June 8, June 22, July 20, and August 24, 1987 correspond to a decrease in soil volumetric moisture (Figure-9). On the Worthing soil for these same sampling dates, the volumetric moisture also decreased. The soil on these tillage plots (Worthing soil) was wetter and therefore probably did not dry out as quickly. Conditions for denitrification had been optimum, so that the organisms could have utilized the nitrate and glucose faster than mineralization could occur. The no and ridge tillage plots had a higher volumetric moisture content than the reduced and conventional tillage plots on June 8 and 22, 1987. The reduced tillage Worthing plots had the highest moisture content on July 20 (34%) and the no tilled plots on August 24, 1987 (26%).

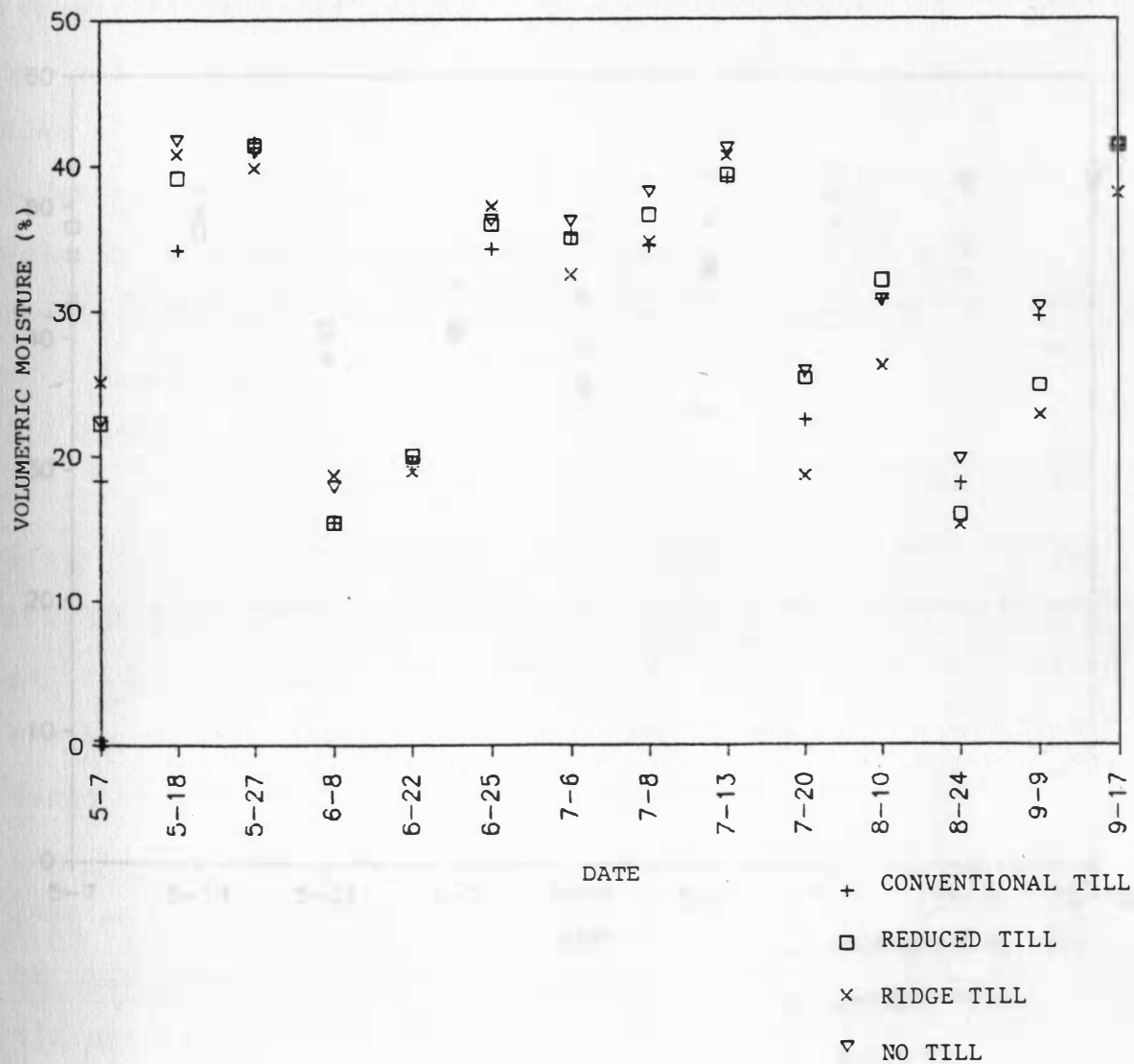


Figure 9. Volumetric moisture on the Beadle Soil, 1987. Each plotted point is an average of three observations.



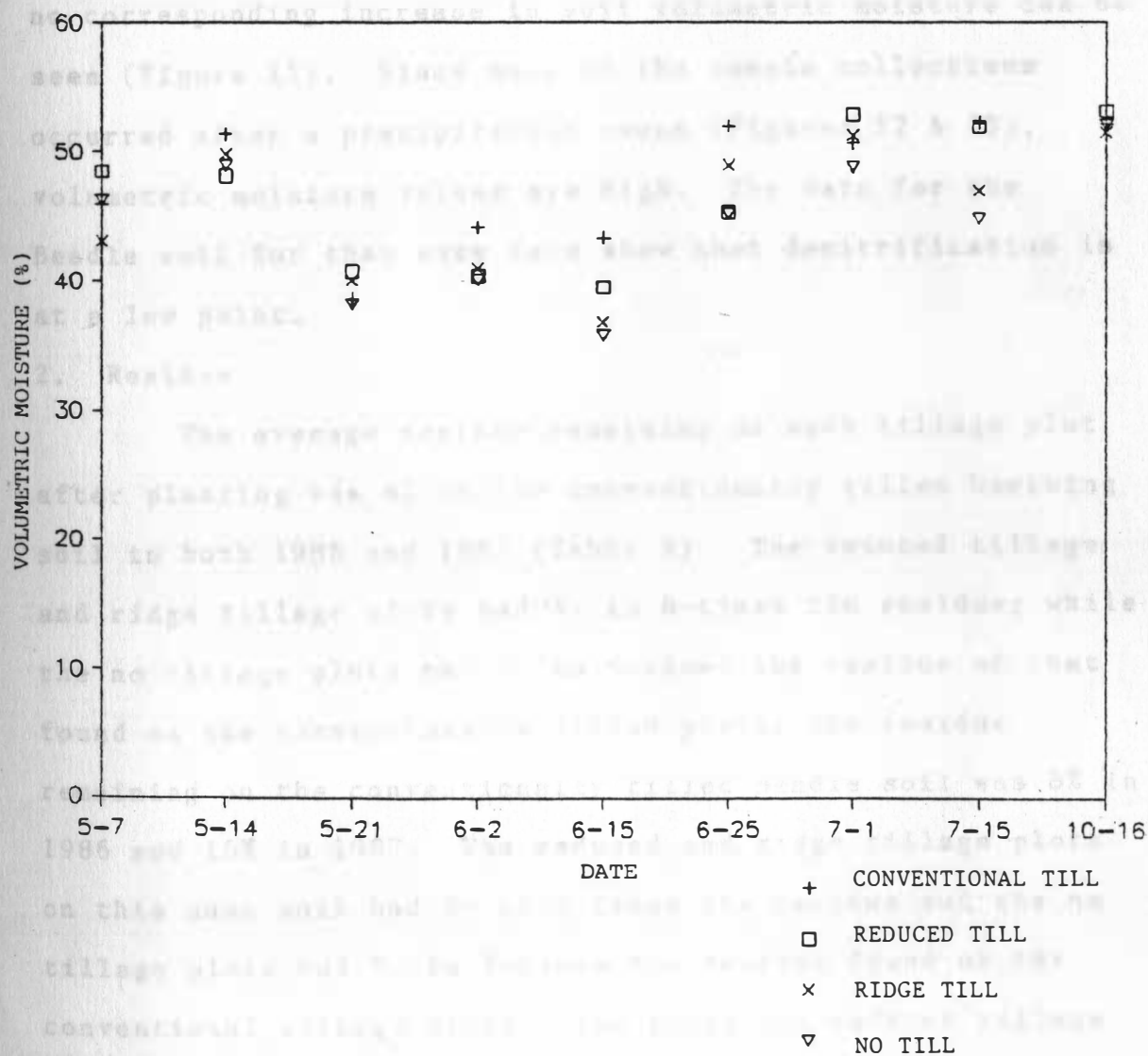


Figure 10. Volumetric moisture on the Worthing Soil, 1986.  
Each plotted point is an average of three observations.

High values of volumetric moisture in the sample did not correspond to periods of high denitrification. Notice that on July 6, 1987 there is a peak in denitrification (Figure 7) on three of the four Worthing tillage plots, yet no corresponding increase in soil volumetric moisture can be seen (Figure 11). Since many of the sample collections occurred after a precipitation event (Figures 12 & 13), volumetric moisture values are high. The data for the Beadle soil for that same date show that denitrification is at a low point.

## 2. Residue

The average residue remaining on each tillage plot after planting was 6% on the conventionally tilled Worthing soil in both 1986 and 1987 (Table 2). The reduced tillage and ridge tillage plots had 4- to 6-times the residue; while the no tillage plots had 6- to 8-times the residue of that found on the conventionally tilled plots. The residue remaining on the conventionally tilled Beadle soil was 8% in 1986 and 15% in 1987. The reduced and ridge tillage plots on this same soil had 4- to 5-times the residue and the no tillage plots had 5- to 7-times the residue found on the conventional tillage plots. The ridge and reduced tillage management produced about the same amount of residue for each year, yet the denitrification that would be expected from each should be different. The reduced tillage method

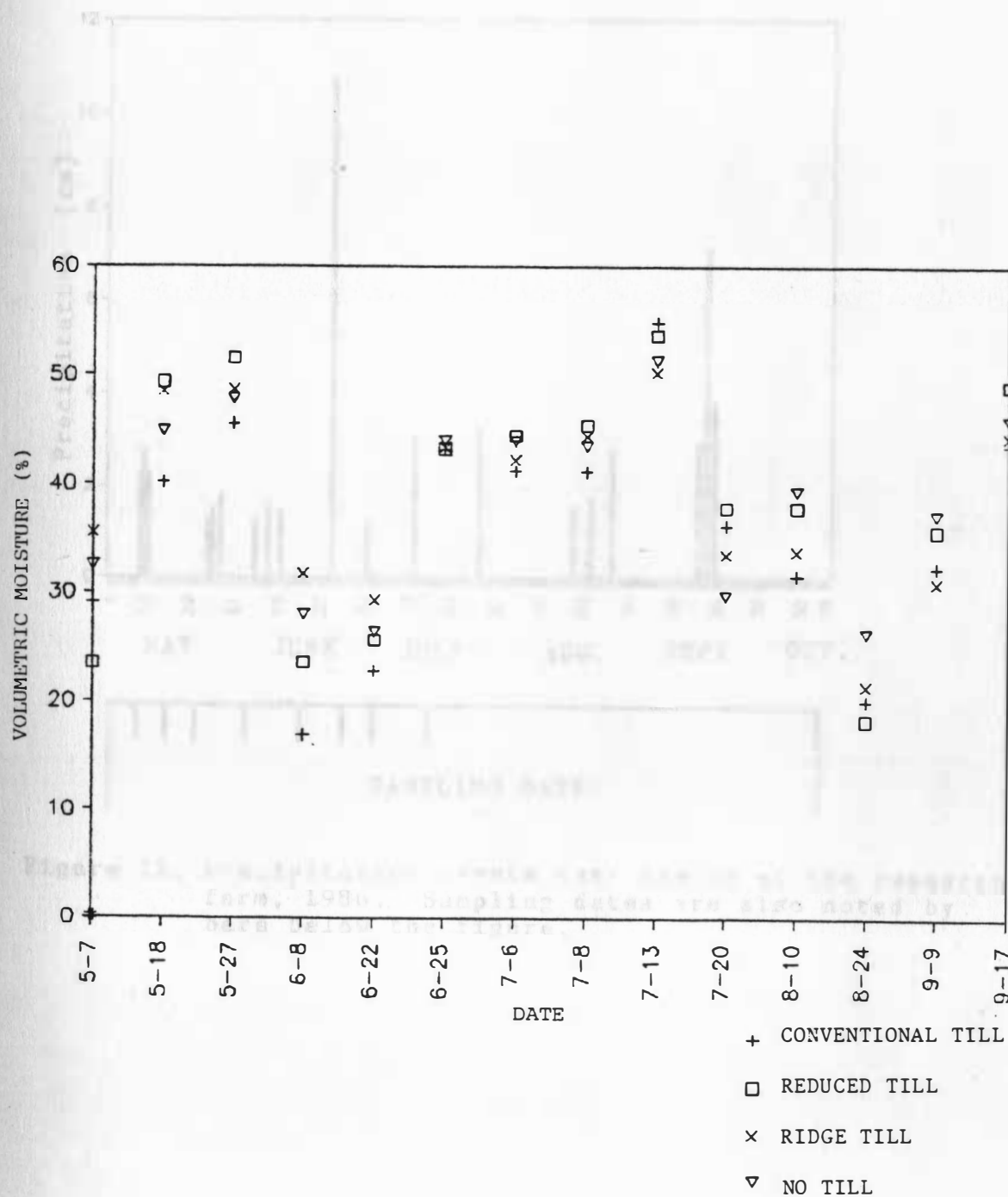


Figure 11. Volumetric moisture on the Worthing Soil, 1987. Each plotted point is an average of three observations.

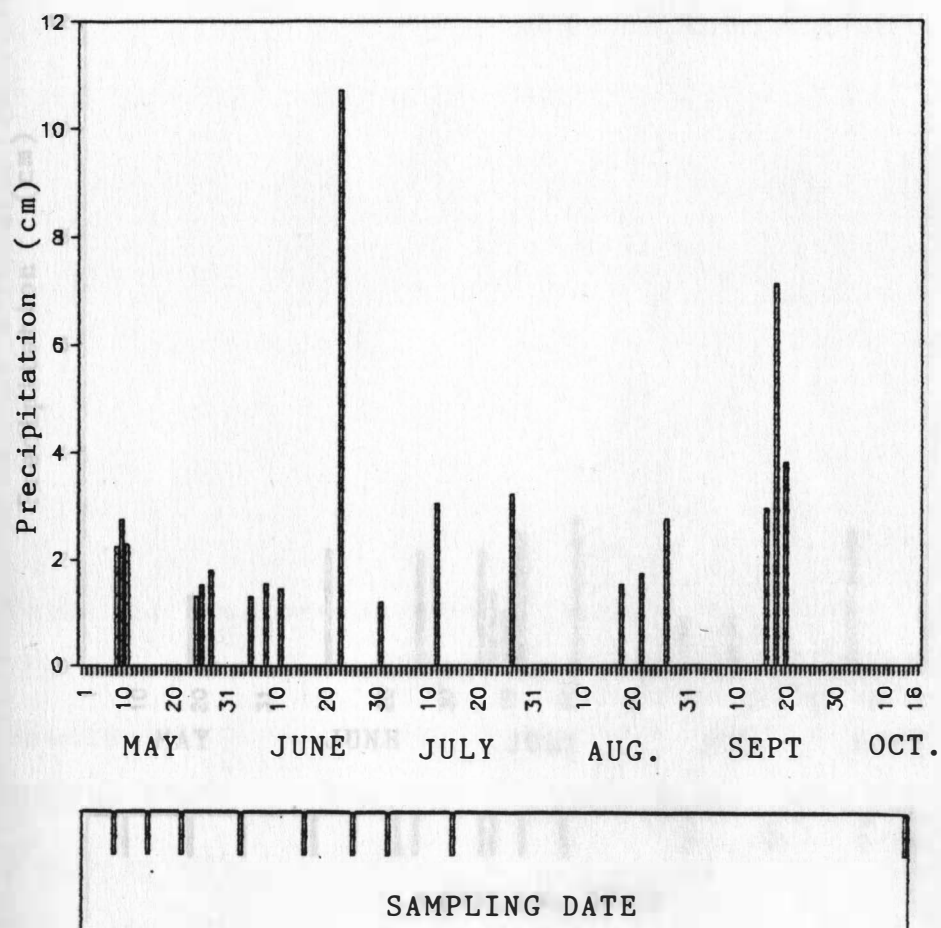


Figure 12. Precipitation events over one cm at the research farm, 1986. Sampling dates are also noted by bars below the figure.

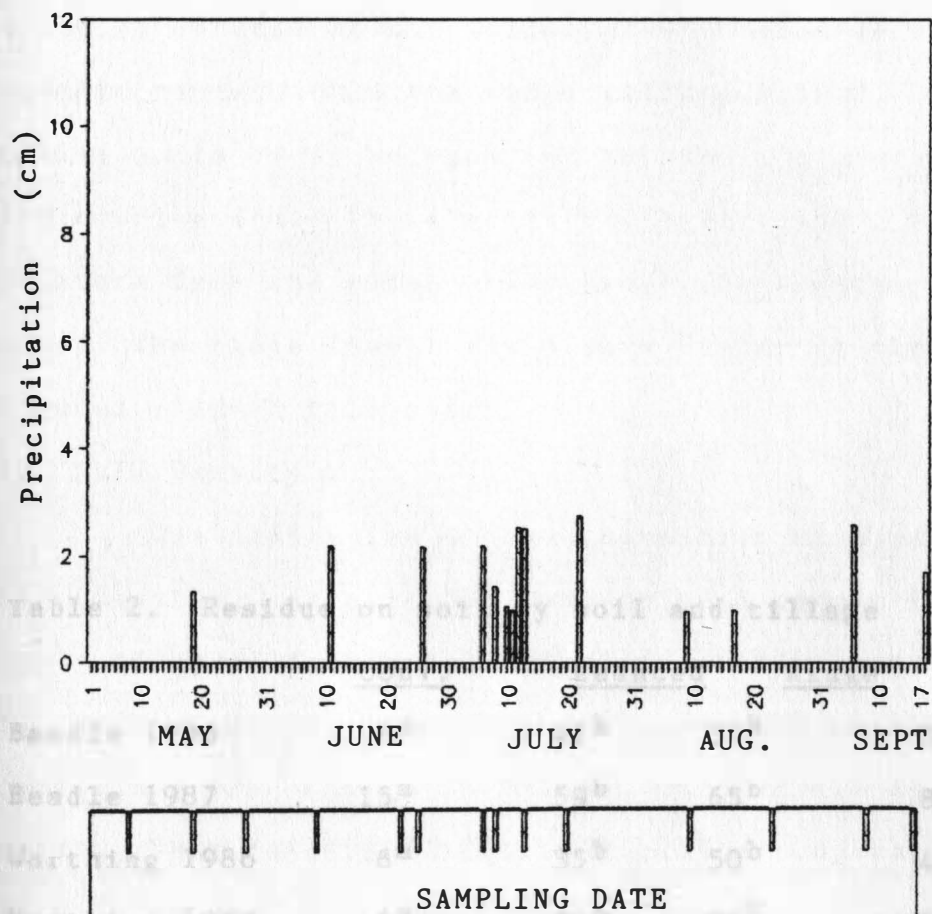


Figure 13. Precipitation events over one cm at the research farm, 1987. Sampling dates are also noted by bars below the figure.





would be aerated by the chisel plow, therefore creating more aerobic centers than the ridge tillage method. The ridge tilled soils could be expected to have a higher rate of denitrification. These soils would act like a wick, pulling moisture from the moist areas under the residue between the rows. The ridge itself would warm faster in the flat and covered reduced till soils.

### 3. Bulk Density

One indication of the compaction of a soil is its bulk density. As the bulk density of a sample increases, the void space decreases, therefore, the amount of oxygen for use by aerobic organisms decreases and anaerobic conditions can occur at a quicker pace with a precipitation event. Bulk density of denitrification samples, when used as a dependent variable, had significant differences between soils, years, and in the soils by tillage interaction. This indicates that the tillage methods on the two soils produced different bulk densities and that the bulk densities on the two soils and in the two years were different. On the Beadle soil, the bulk density value averages for the two years ranged from 1.18 to 1.19 for each of the tillage treatments (Figure 14). In contrast to these values, the average bulk density values for the Worthing soil ranged from 1.07 and 1.08 on the conventionally tilled and reduced tilled soils to 1.13 and 1.16 on the ridge tilled and no tilled soil

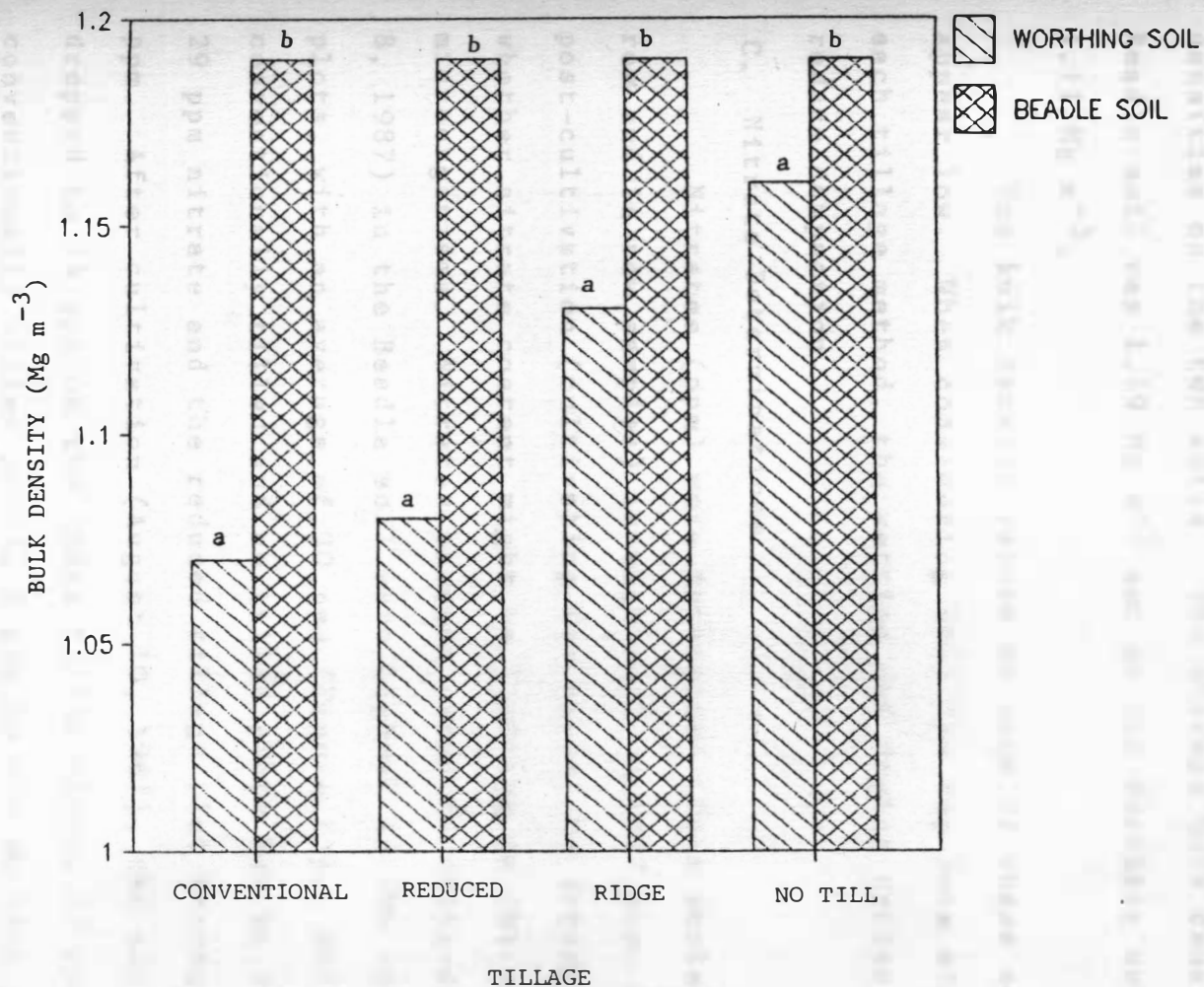


Figure 14. Average (1986 and 1987) bulk density values for the Worthing and Beadle soils. Each plotted bar is an average of 69 observations. Superscripts of different letters indicate differences significant at the 0.05 level.

plots. Those tillage plots which were mixed by tillage the most had the lower bulk density on the Worthing soil. There were significant (.05) differences between the average bulk densities on the two soils. The average bulk density on the Beadle soil was  $1.19 \text{ Mg m}^{-3}$  and on the Worthing soil was  $1.11 \text{ Mg m}^{-3}$ .

The bulk density values on each of these soils appear low. When considering only the top 5-cm of soil from each tillage method, the wetting and drying cycles tend to reduce compaction.

#### C. Nitrate Determinations

Nitrates (ppm) were determined (from pooled between row and in row samples) in each soil in 1987 pre- and post-cultivation (cultivating for weeds) to determine whether nitrate content might be limiting to the soil microorganisms. Nitrates present prior to cultivation (July 8, 1987) in the Beadle soil were highest in the ridge tilled plots, with an average of 30 ppm (Figure 15). Both the conventionally tilled and no tilled plots had an average of 29 ppm nitrate and the reduced tillage plot average was 20 ppm. After cultivation (August 10, 1987), the nitrate level dropped to 14 ppm on the ridge tilled plots, 12 ppm on the conventionally tilled plots, 8 ppm on the no till plots and 6 ppm on the reduced till plots. On the Worthing soil

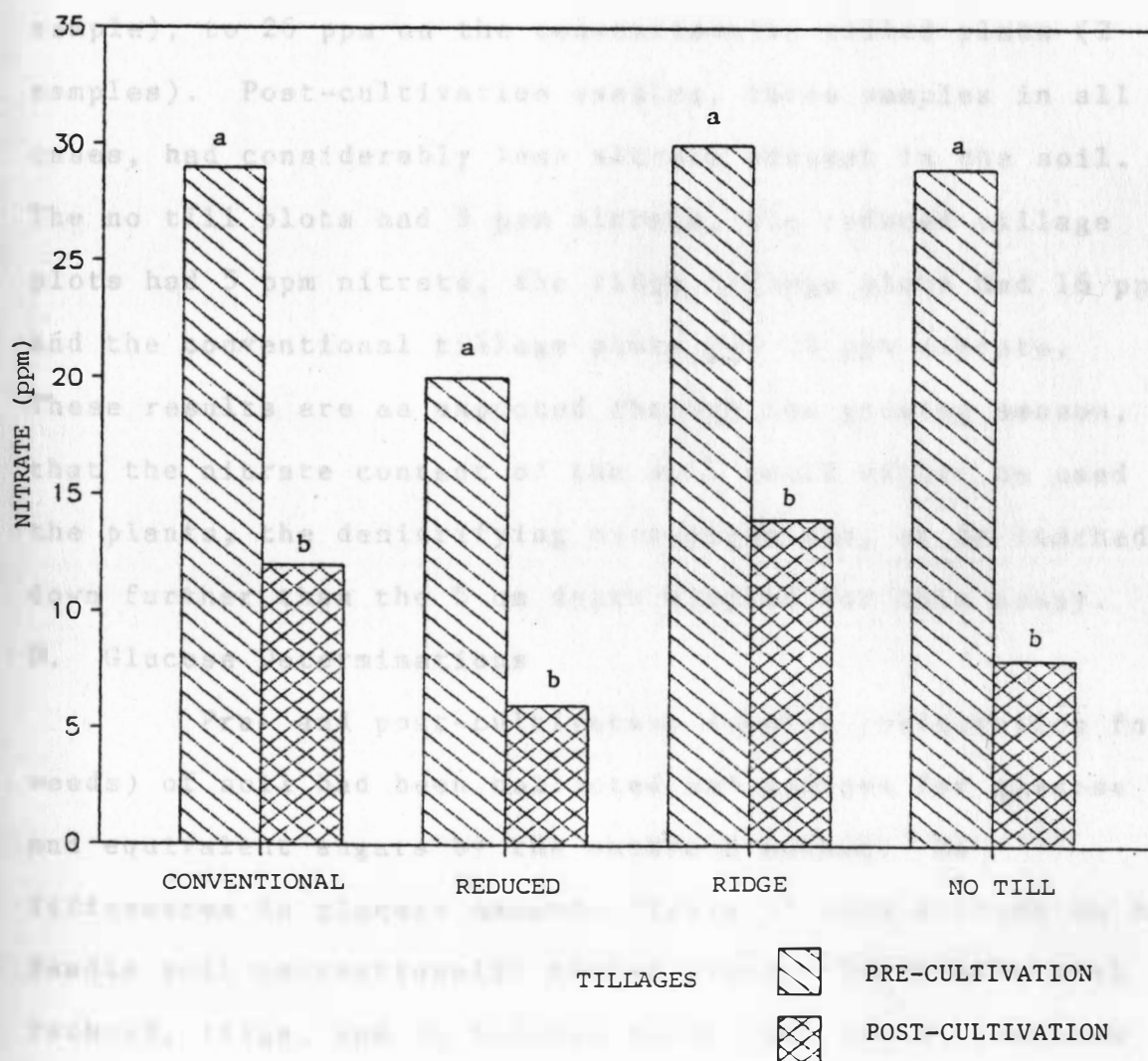


Figure 15. Nitrate amounts on the Beadle Soil pre- and post-cultivation, 1987. Each plotted point is an average of three observations. Superscripts of different letters indicate differences significant at the 0.05 level.



(Figure 16), there was an incomplete set of samples among the pre-tillage samples. The pre-cultivation nitrate content on the Worthing soil ranged from 61 ppm on the no till plots (average of 2 samples), 45 ppm on the reduced tillage plots (2 samples), 40 ppm on the ridge till plots (1 sample), to 26 ppm on the conventionally tilled plots (2 samples). Post-cultivation samples, three samples in all cases, had considerably less nitrate present in the soil. The no till plots had 5 ppm nitrate, the reduced tillage plots had 5 ppm nitrate, the ridge tillage plots had 16 ppm and the conventional tillage plots had 10 ppm nitrate. These results are as expected through the growing season, that the nitrate content of the soil would either be used by the plants, the denitrifying microorganisms, or be leached down further than the 5 cm depth sampled for this assay.

#### D. Glucose Determinations

Pre- and post-cultivation samples (cultivation for weeds) of soil had been collected and assayed for glucose and equivalent sugars by the anthrone method. No differences in glucose amounts (Table 3) were evident on the Beadle soil conventionally tilled plots. The Beadle soil reduced, ridge, and no tillage plots each had an increase in glucose equivalents of from 40 to 80  $\mu\text{g g}^{-1}$  soil. Glucose amounts in the Worthing soil conventionally tilled plots decreased from 348- to 287- $\mu\text{g glucose g}^{-1}$  soil. The reduced

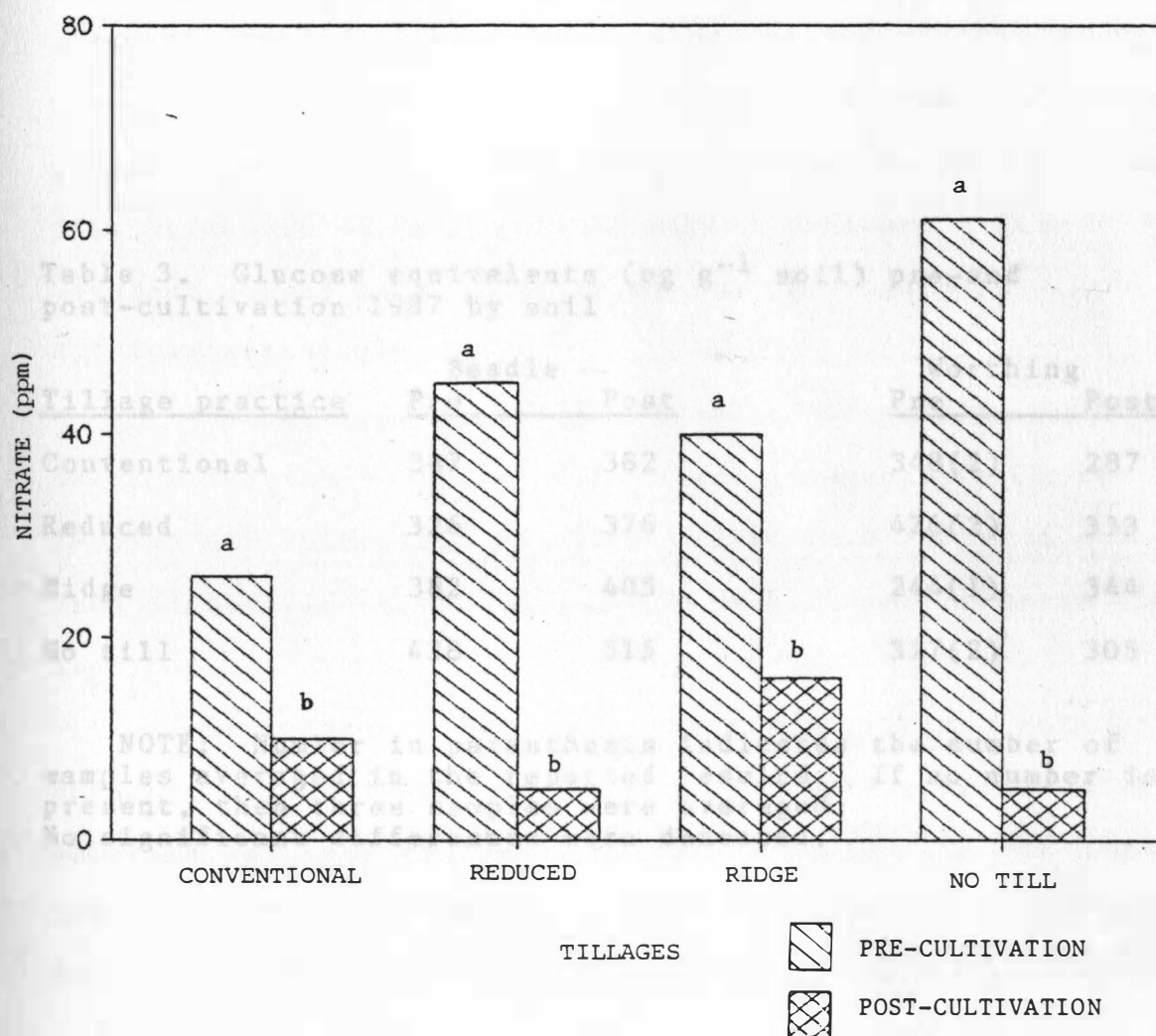


Figure 16. Nitrate amounts on the Worthing Soil pre- and post-cultivation, 1987. Each plotted point is an average of three observations. Superscripts of different letters indicate differences significant at the 0.05 level.

Table 3. Glucose equivalents ( $\mu\text{g g}^{-1}$  soil) pre-and post-cultivation 1987 by soil

<u>Tillage practice</u>	<u>Beadle</u>		<u>Worthing</u>	
	<u>Pre</u>	<u>Post</u>	<u>Pre</u>	<u>Post</u>
Conventional	387	382	348(2)	287
Reduced	326	376	476(2)	333
Ridge	382	405	244(1)	344
No till	438	515	317(2)	305

NOTE: Number in paranthesis indicates the number of samples averaged in the reported results. If no number is present, then three samples were averaged. No significant differences were detected.

tillage plots had a decrease from 476- to 333-ug glucose and the no till plots had a slight decrease from 317- to 305-ug glucose  $\text{g}^{-1}$  soil. Only the ridge tillage plots had evidence of an increase in glucose from 244 to 334 ug  $\text{g}^{-1}$  soil. There are no statistical differences between pre- and post-cultivation glucose amounts or between glucose amounts present in the soil under each tillage treatment. There is probably sufficient organic matter present in each soil so that breakdown of this organic matter produces a fairly constant supply of glucose or equivalent sugars.

#### E. Statistical Analyses

To determine possible causes of the higher denitrification in these particular tillage plots, bulk density and volumetric moisture values were analyzed and used as covariants with kilograms of nitrogen released per day.

Significant differences were found by analysis of variance for soil and for the soil by year interaction when moisture was used as the dependent variable. On the Beadle soil, the 1986-1987 average volumetric moisture was 30.5% and, on the Worthing soil, it was 42.0%. When averaged over soils, 1986 had an average volumetric moisture of 38.5%, where in 1987, the average was 34.0%. Yet, denitrification was not significantly higher in 1986. The higher moisture content probably was due to the increased sampling after

precipitation events.

The volumetric moisture content (average for each rep of each tillage treatment) was higher in the Worthing soil (46.1%) than in the Beadle soil (30.8%) in 1986 due to the landscape position of the two soils. The Worthing soil average moisture content on all sites ranged between 43.8 and 49.5% in 1986 while the average moisture content on the Beadle soil ranged between 29.3 and 32.5%. The soil by year interaction of moisture as analyzed by the statistical package was determined to be very highly significant ( $p < 0.001$ ). This means that each soil varied by year with respect to volumetric moisture content. The differences in 1986 are explained above. In the final year of the study (1987) there were less dramatic differences in moisture with the average on the Worthing soil being 37.8% and on the Beadle soil, 30.1%. Only a 7- to 13-% difference between the average moistures was determined. These same sites had average volumetric moisture contents ranging between 34.9 and 40.8% on the Worthing and 27.5 and 33.7% on the Beadle soil. Figure 17 shows graphically, the average volumetric moisture summed over replications and years for each tillage treatment. Volumetric moisture values on the Beadle soil ranged from 29.7% for the ridge tilled soil to 31.6% for the no till managed soil. In contrast to these values, on the Worthing soil the volumetric moisture values ranged from



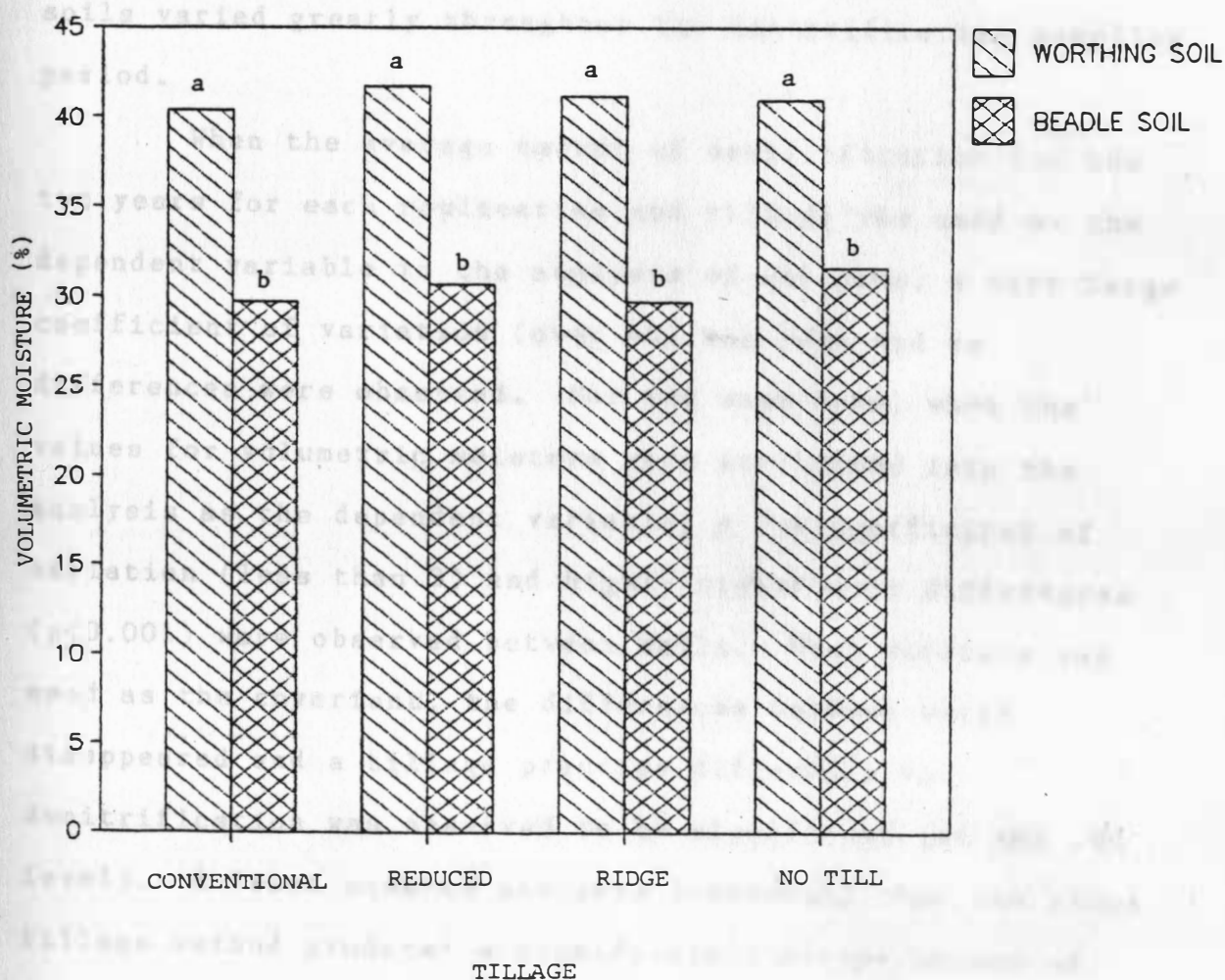


Figure 17. Average volumetric moisture (1986 and 1987) for the four tillage systems on the Beadle and Worthing soils. Each plotted bar is an average of 69 observations. Superscripts of different letters indicate differences significant at the 0.05 level.

40.4 to 41.7% on the conventional and reduced tillage plots respectively. When these values of volumetric moisture were averaged for the two years and analyzed using the SAS package, once again, highly significant differences were seen. This indicates that the moisture status of the two soils varied greatly throughout the denitrification sampling period.

When the average amount of denitrification for the two years for each replication and tillage was used as the dependent variable in the analysis of variance, a very large coefficient of variation (over 40) was seen and no differences were observed. For the same data, when the values for volumetric moisture were introduced into the analysis as the dependent variable, a low coefficient of variation (less than 3) and highly significant differences ( $p < 0.001$ ) were observed between soils. When moisture was used as the covariant, the differences between soils disappeared and a tillage practice difference in denitrification was observed to be significant (at the .05 level). A least squares analysis determined that the ridge tillage method produced a significantly higher amount of denitrification than did the reduced, conventional, or no tillage methods of soil management (Figure 18). These other three tillage methods did not differ in respect to the amount of denitrification occurring on them. The adjusted

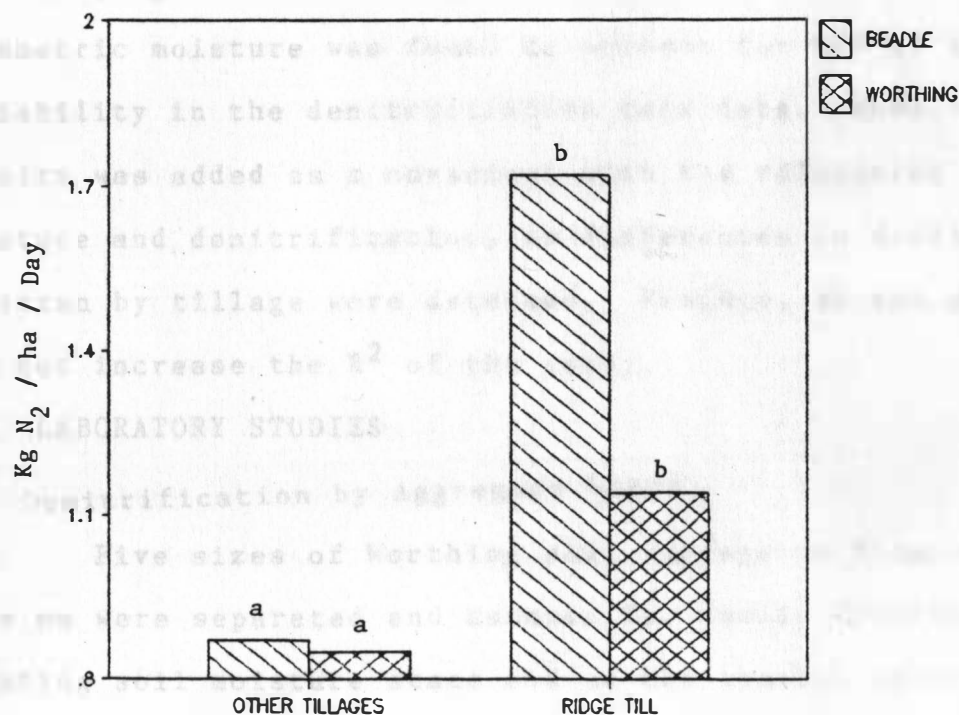


Figure 18. Comparison of denitrification rate in ridge tillage plots versus the average of the three other tillage methods. Years are combined. For the ridge tillage method, 69 observations are averaged. For the other tillage methods, 207 observations are averaged. Superscripts of different letters indicate differences significant at the 0.05 level.

denitrification occurring on the Beadle soil ridge tillage plots was 1.14-kg nitrogen loss  $\text{ha}^{-1} \text{ day}^{-1}$  and the average of that occurring on the other three tillage plots was 0.85-kg loss  $\text{ha}^{-1} \text{ day}^{-1}$ . On the Worthing soil, losses on the ridge tilled soil were 1.72-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$  and 0.87-kg nitrogen loss  $\text{ha}^{-1} \text{ day}^{-1}$  on the other three tillage systems (Figure 17). Using this covariant analysis, volumetric moisture was found to account for 67% of the variability in the denitrification rate data. When bulk density was added as a covariant with the volumetric moisture and denitrification, no differences in denitrification by tillage were detected. Residue, pH and glucose did not increase the  $R^2$  of the assay.

## II. LABORATORY STUDIES

### A. Denitrification by Aggregate Sizes

Five sizes of Worthing soil aggregates from one to nine mm were separated and assayed for denitrification in an existing soil moisture state and on the tension table at 50 cm tension. The mean denitrification occurring in the less than one mm size aggregates was 0.17-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$ . For the 1- to 2-mm size, 0.32 kg; for the 2- to 4.75-mm size, 0.21 kg for the 4.75- to 6.36-mm size, 0.21 and for the 6.36- to 9.5-cm size, 0.18-kg nitrogen  $\text{ha}^{-1} \text{ day}^{-1}$  was denitrified from the soil. There were no significant differences on the tillage treatment level or between the

reps. This relates to what was determined by Sexstone et al. (1985b), who stated that aggregate anaerobic sites were not the only factor involved in denitrification rate.

#### B. Equilibration Time on the Tension Table

This curve was developed to determine which depth of tension to use to closely approximate field gravimetric moisture conditions. An experiment to determine the amount of time to leave the soil on the tension table to equilibrate soil moisture within the sample produced the following results: Soil nitrous oxide production quickly reached a plateau, going from 175-ppm to 0-ppm nitrous oxide production in the study period of 72 hours. Within 24 hours, samples of soil had a stable moisture content of 45% gravimetrically. This was equivalent to the moisture content after 72 hours of incubation. At the same time, nitrous oxide production appeared to steadily decrease after the sample was on the tension table for six hours. Soil samples left on the tension table longer than six hours exhibited a decreasing ability to denitrify. This could in part be due to the high moisture content of the soil when the tension table was flooded, which could provide optimum conditions for denitrification. With time, the moisture content and the amount of nitrate in the soil would decrease. The time of 24 hours was selected as a reasonable period for equilibration on the tension table after which



the soil denitrifiers would still produce enough nitrous oxide for analysis.

When a Worthing soil sample was placed upon the tension table and the tension was adjusted to 25, 50, and 75 cm (0.025, 0.05, and 0.075 bar), a partial water retention curve was obtained which showed the moisture content at each of these tensions (Figure 19). Values for 0.33 and 1.0 bar moisture were obtained by the use of a pressure plate apparatus. The values obtained by the use of the sand tension table appear to fit the upper end of the curve in a reasonable manner.

#### C. Glucose and Nitrate Additions

The 2 to 4.75 mm size fraction of the ridge till plots on the Worthing soil in replication 3 were used to determine the effect of addition of nitrate and glucose upon denitrification. These soil samples were placed upon the tension table at 50 cm tension. When 625 ug glucose alone  $\text{g}^{-1}$  soil was added, nitrous oxide production increased from a background of 2790 ppm to 5100 ppm. As the amount of glucose increased, the denitrification seemed to level off, indicating that at that level of glucose, it is not limiting denitrification. There was 250 ug glucose present in the soil samples prior to glucose additions.

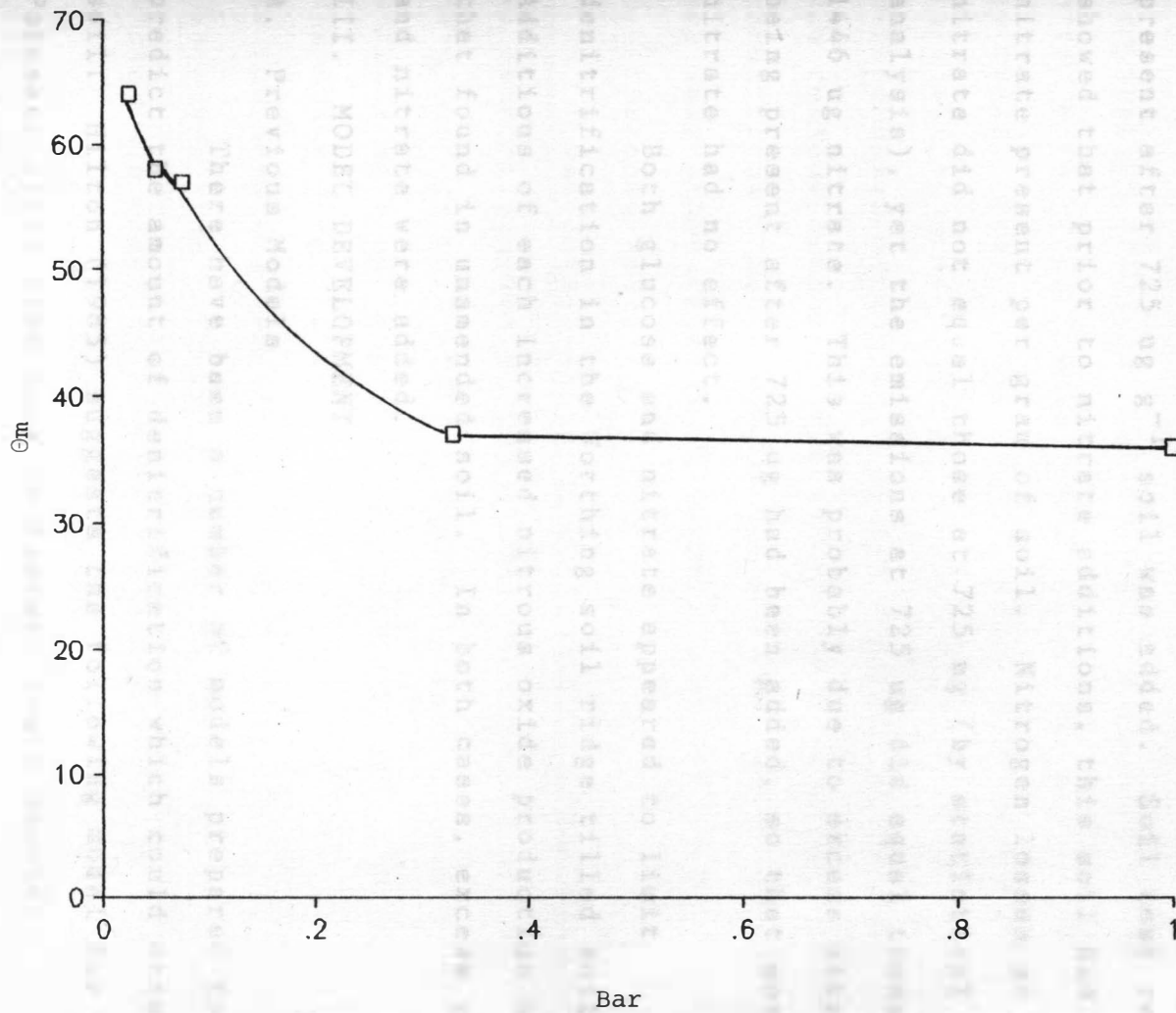


Figure 19. Partial moisture retention curve for the Worthing soil.

As adjusted soil nitrate content increased from 0 to 725 to 1446  $\text{ug g}^{-1}$  soil, the nitrous oxide production increased from 2890 ppm to 5805 ppm and then leveled off to 6260 ppm. This also indicates that sufficient nitrate was present after 725  $\text{ug g}^{-1}$  soil was added. Soil test results showed that prior to nitrate additions, this soil had 15  $\text{ug}$  nitrate present per gram of soil. Nitrogen losses at 0  $\text{ug}$  nitrate did not equal those at 725  $\text{ug}$  (by statistical analysis), yet the emissions at 725  $\text{ug}$  did equal those at 1446  $\text{ug}$  nitrate. This was probably due to excess nitrate being present after 725  $\text{ug}$  had been added, so that more nitrate had no effect.

Both glucose and nitrate appeared to limit denitrification in the Worthing soil ridge tilled soil. Additions of each increased nitrous oxide production over that found in unamended soil. In both cases, excess glucose and nitrate were added.

### III. MODEL DEVELOPMENT

#### A. Previous Models

There have been a number of models prepared to predict the amount of denitrification which could occur in a soil. Hilton (1985) suggests the following model for a Poinsett silty clay loam in Eastern South Dakota:

$Y = -3.9026 + 0.25367(a) - 0.01507(b)$ , where 'a' is soil temperature and 'b' is soil temperature \* days after

rainfall. A second model has a number of different factors:  
 $\hat{Y} = 0.2388 - 0.0867 (c) + 0.00627 (d) + 0.00000865 (e) +$   
 $0.0000148 (f) - 0.226 (g)$ . This model includes 'c' =  
 percent residue cover, 'd' = percent residue cover \* soil  
 temperature ( $^{\circ}\text{C}$ ), 'e' residue cover squared \* number of days  
 after a rainfall \* soil temperature, 'f' = soil water  
 potential (kPa) \* soil temp \* number of days after a  
 rainfall squared, and 'g' = dummy variable (1=injected and  
 0=topdressed nitrogen). Hilton's model relies heavily on  
 the residue factors. His second model includes residue in  
 three terms. Soil temperature and soil water potential are  
 the only other factors involved.

#### B. Proposed Model

Volumetric moisture is the only significant factor  
 in denitrification in the empirical model (Table 4)  
 developed in this study. Residue amount, pH, and bulk  
 density were not significant as covariants. There is a  
 resemblance to Hilton's model in that residue cover relates  
 to the moisture held by the soil. Therefore, as residue  
 increases, the moisture held by the soil also increases,  
 making his model more moisture related than is evident.

Pre- and post-cultivation glucose equivalents and  
 nitrate concentrations were not added to this analysis since  
 these measurements were only collected for one year.

# IV. REGRESSION ANALYSIS OF THE DATA

Regression analysis was performed on the data for each of the three systems. The results are shown in Figures 23 through 25. The data were first plotted as a daily loss and the loss was then plotted as a function of volumetric moisture. The results of the regression analysis are shown in Table 4. The regression equation for the loss of nitrogen is  $\hat{Y} = -4.379 + 0.196(a)$  where  $\hat{Y}$  is the loss of nitrogen in kg per day and  $a$  is the volumetric moisture in percent.

Table 4. Summary of proposed model

<u>dependent variable</u>	<u>equation</u>
kg nitrogen lost per day	$\hat{Y} = -4.379 + 0.196(a)$

$a$  = volumetric moisture (xx.x)

$$r^2 = 0.67$$

NOTE: Use of this equation should be limited to volumetric moisture values in the range of 15% to 55%. Values outside this range were not studied and might produce erroneous results.



#### IV. SIGNIFICANT LOSSES DUE TO DENITRIFICATION

Estimated kg nitrogen lost per year on each tillage system was derived by viewing the data in Figures 20 through 23. The peak amounts were entered as a daily loss and the next two to three days, the peak amount was decreased to half and then one fourth of the original value. When a steady trend of above zero denitrification is seen, that value is taken by the number of days which make up the trend to create an estimated denitrification value for each tillage system. The values in this table (Table 5) could be used with the values in Table 6 to prepare a nitrogen budget. A sample budget (Table 7) shows that on the Beadle and Worthing Ridge tillage plots, the denitrification amount, when added to the loss of nitrogen in the grain compare favorably with estimated nitrogen inputs. Where 211 pounds of nitrogen is added in 1987 to the Beadle soil ridge till plots (through fertilization, original nitrate content, and organic matter mineralization) losses of 217 pounds can be estimated. This demonstrates that the denitrification value arrived at is reasonable, since the estimated nitrogen loss closely approximates the nitrogen additions.

Figure 20. Denitrification by Julian date, 1986 for each tillage system on the Beadle soil. Three observations are averaged in each data point.

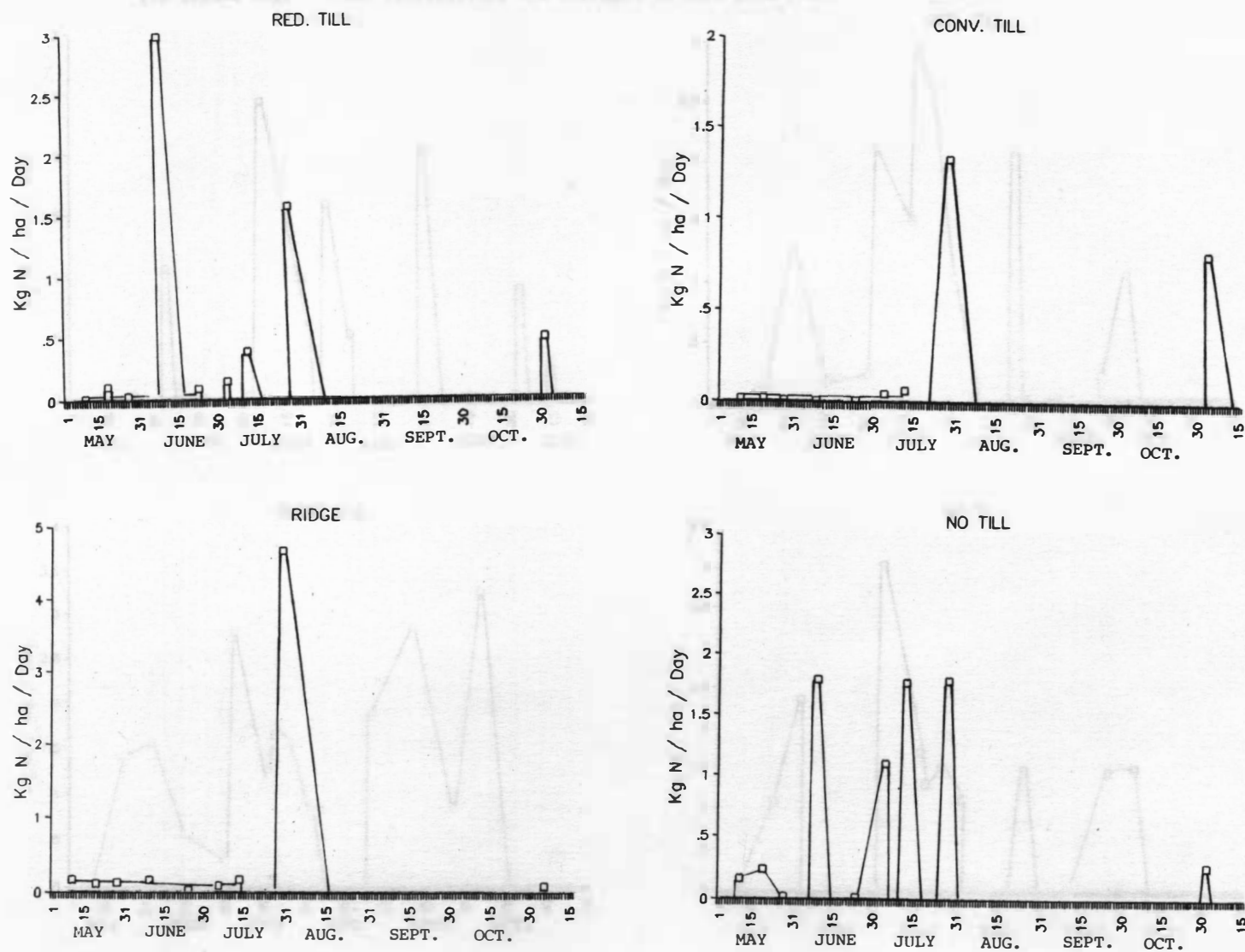


Figure 21. Denitrification by Julian date, 1987 for each tillage system on the Beadle soil. Three observations are averaged in each data point.

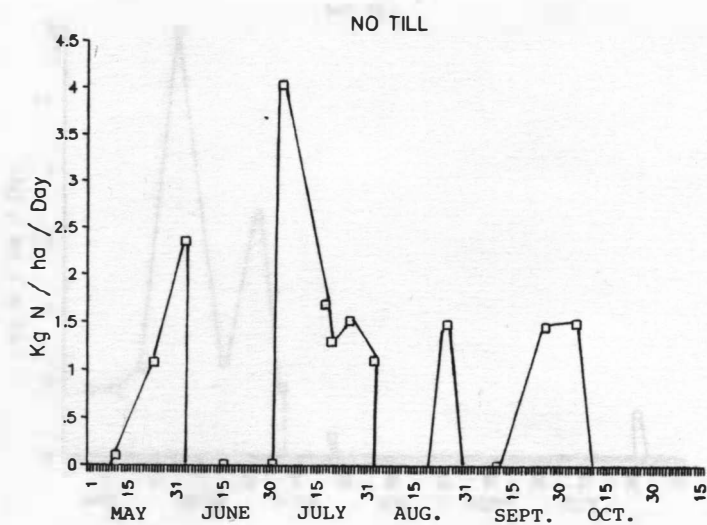
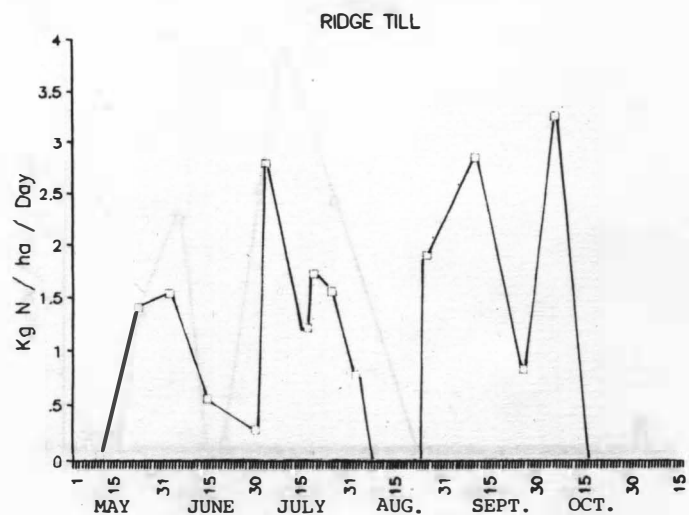
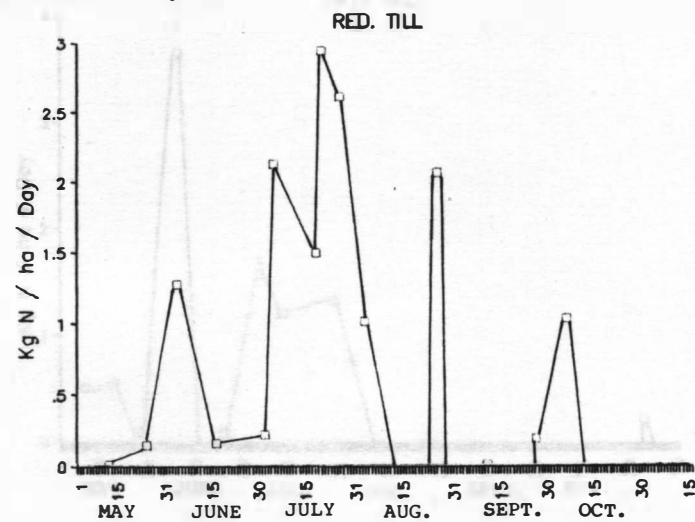
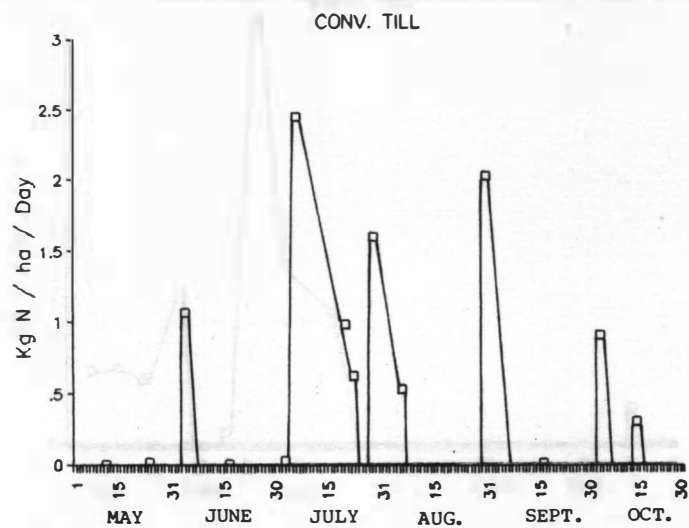


Figure 22. Denitrification by Julian date, 1986 for each tillage system on the Worthing soil. Three observations are averaged in each data point.

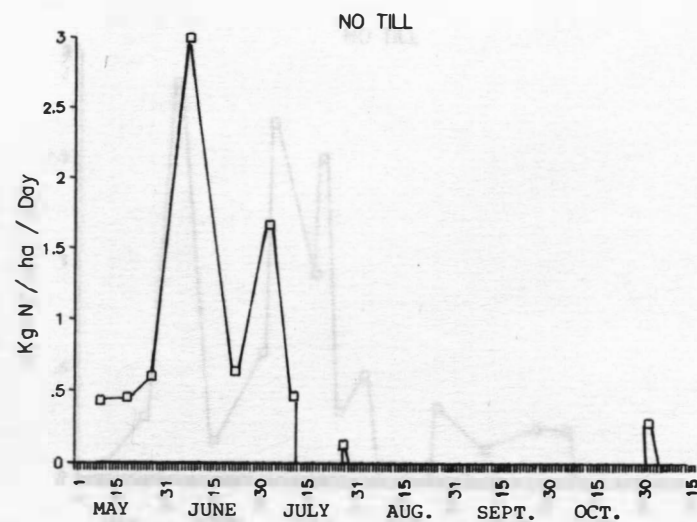
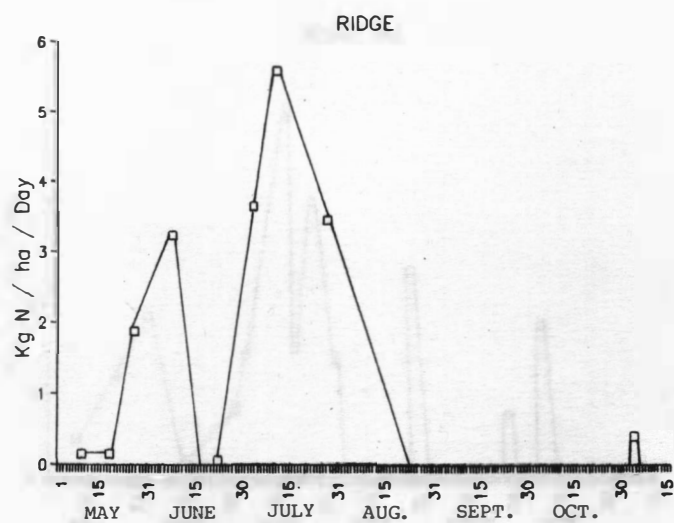
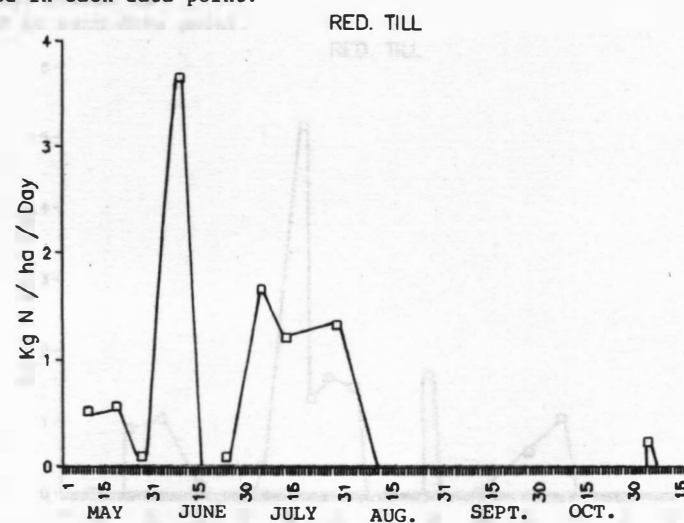
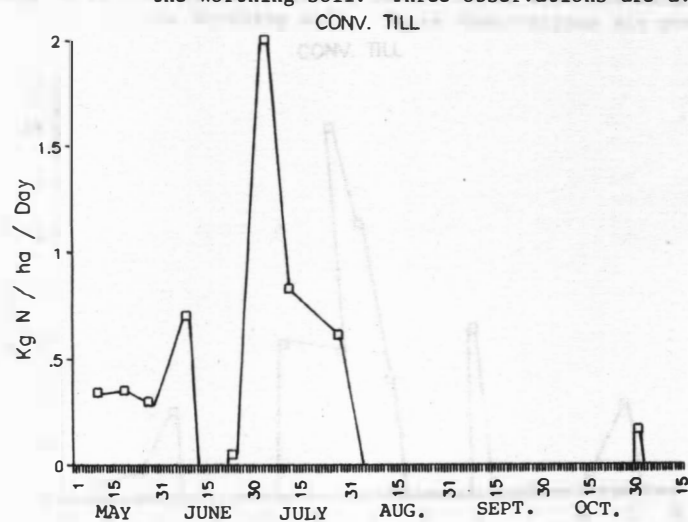


Figure 23. Denitrification by Julian date, 1987 for each tillage system on the Worthing soil. Three observations are averaged in each data point.

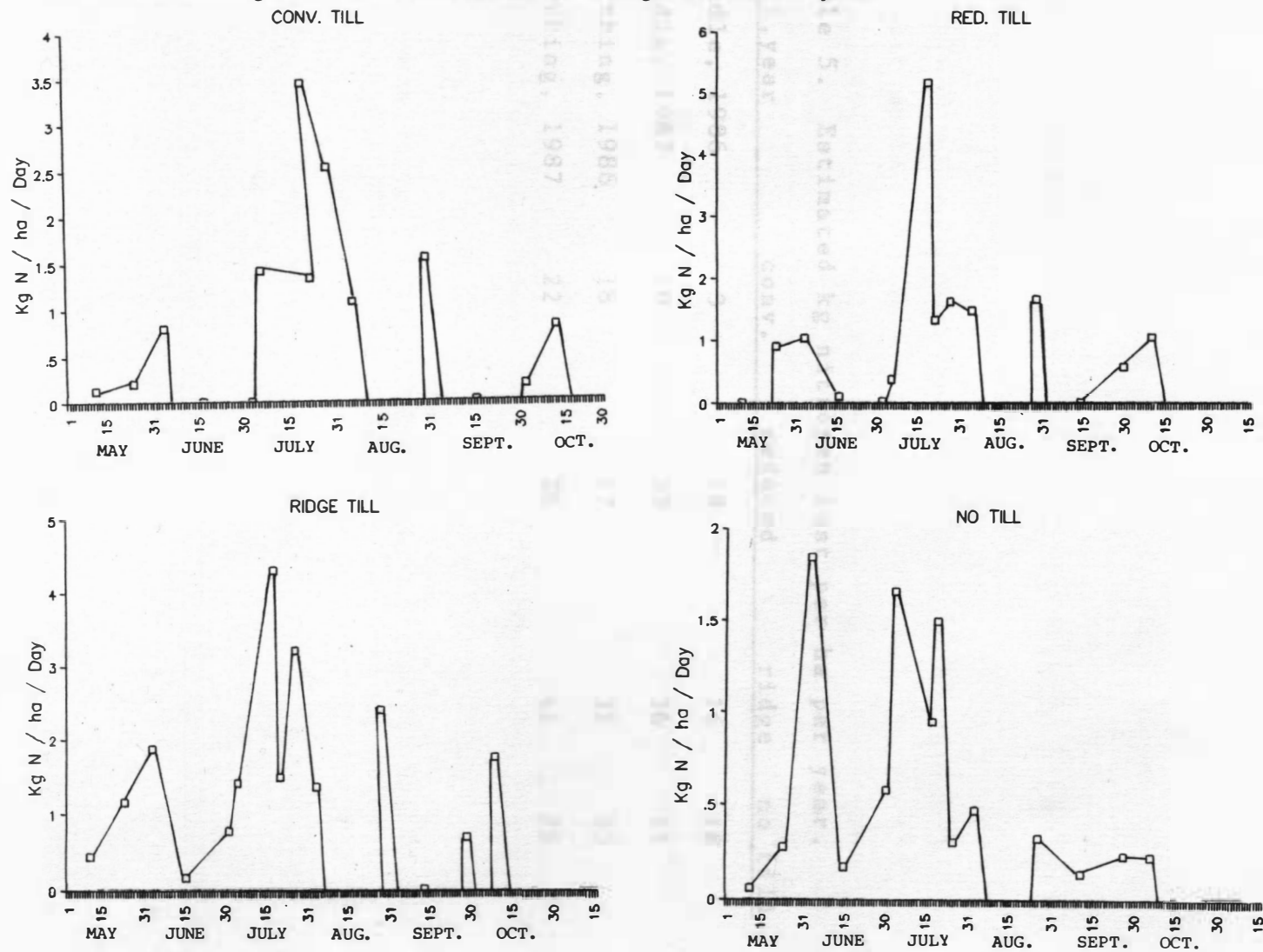




Table 5. Estimated kg nitrogen lost per ha per year.

<u>soil, year</u>	<u>conv.</u>	<u>reduced</u>	<u>ridge</u>	<u>no till</u>
Beadle, 1986	3	18	16	18
Beadle, 1987	19	35	36	31
Worthing, 1986	18	17	31	23
Worthing, 1987	22	26	41	25

Table 6. Factors needed to prepare a nitrogen budget.

<u>Beadle Soil</u>	<u>conv.</u>	<u>reduced</u>	<u>ridge</u>	<u>no till</u>
Organic matter	3.5	3.2	4.1	4.0
Nitrate*	19	22	19	11
Yield	180	141	144	143
<u>Worthing Soil</u>	<u>conv.</u>	<u>reduced</u>	<u>ridge</u>	<u>no till</u>
Organic matter	4.4	4.0	4.4	3.9
Nitrate*	13	11	24	7
Yield	106	111	116	119

\* Nitrate content here is that found in the top six inches prior to planting.

Table 7. Sample nitrogen budget calculations.

Beadle soil, ridge tilled: yield = 144 bu a<sup>-1</sup>, OM=4.1%,  
nitrates = 19, denitrification = 36 kg n ha<sup>-1</sup> day<sup>-1</sup>

## INPUTS:

fertilizer N = 110 lbs  
nitrate (top 6") = 19 lbs  
mineralized n = 82 lbs

## LOSSES:

Corn\* 189 lbs  
denit.\*\*\* 28 lbs

## TOTALS:

211 lbs.

217 lbs.

\* 189 = (144\*1.45)-20, where 144 = yield and 1.45 = nitrogen factor

\*\* 82 = .041 [organic matter content] \* 2\*10<sup>6</sup> \* .05 \* .02  
.05 is the amount of nitrogen in organic matter and  
.02 is the mineralization rate of the organic matter.

\*\*\* 28 = 36 kg nitrogen lost ha<sup>-1</sup> year<sup>-1</sup> \* 0.892 = lbs a<sup>-1</sup>

Worthing soil, ridge tilled: Yield = 116 bu a<sup>-1</sup>, OM = 4.4,  
nitrates = 24 lbs a<sup>-1</sup>, estimated kg nitrogen lost yr<sup>-1</sup> = 41.

## INPUTS:

fertilizer N = 110 lbs  
nitrate (top 6") = 24 lbs  
mineralized n = 88 lbs

## LOSSES:

Corn 148 lbs  
denit 36 lbs

## TOTALS:

222 lbs

184 lbs

## SUMMARY

Denitrification is an important microbial process where, under anaerobic conditions, bacteria utilize nitrates present in soil as terminal electron acceptors. Nitrogen gases ( $N_2$  or  $N_2O$ ) are exported to the atmosphere as a result of this reduction. This process should be enhanced in water saturated soils in which excess nitrates and carbohydrates are present.

Today, denitrification is occurring at a much higher rate than it did 100 years ago. Reasons for this are that nitrogen fertilizer usage has increased dramatically and many grasslands and native prairies have been converted to row crop agriculture. Under grass vegetation, nitrates are effectively utilized by the plant and precipitation is intercepted by the plants, thus providing less favorable conditions for denitrification. Rosswall (1976) has suggested that increased losses of nitrous oxide to the atmosphere have decreased the amount of ozone surrounding the earth, thereby leading to an increased greenhouse effect.

A study designed to investigate denitrification was based on several hypotheses: Denitrification should be greater in the poorly drained soils due to the higher moisture contents. Less disturbed tillage systems would have higher denitrification rates than the conventional or

reduced tillage plots. Surface soil 'glucose equivalents' regulate denitrification and differ between tillage systems. Glucose additions to the soil would increase denitrification. Soil aggregate size would effect denitrification by allowing for a greater soil:aeration ratio.

Many farms in Eastern South Dakota include both well and poorly drained soils. Rates of denitrification on these environments are not known. Two soils in a landscape of the USDA tillage research farm near Madison, South Dakota were selected for this study. Existing plots (established in 1985) of four tillage systems (conventional tillage, reduced tillage (chisel plow-disk), ridge tillage, and no tillage) under continuous corn were used for the two years of this study. Denitrification was monitored on three replications of each tillage treatment by monitoring  $N_2$  /  $N_2O$  gas evolved from soil periodically throughout the two growing seasons (April, 1986 - October, 1986 and May, 1987 - September, 1987). Volumetric moisture and bulk density of these samples was correlated to denitrification. Carbohydrate (glucose) and nitrate amendments were added to representative soils under controlled conditions.

Of the two soil types studied during 1986, the Worthing soil had a higher volumetric moisture content, averaging 46.1 percent, than the Beadle soil, which averaged only 30.8%. This year had been an exceptionally wet year



with the farm receiving 81.7 cm of precipitation between April and September. The Worthing soil in 1987 averaged 37.8% volumetric moisture and the Beadle soil, 30.1%. In 1987, with a more typical precipitation pattern (37.7-cm precipitation), the Worthing soil volumetric moisture average decreased by almost 10% but the Beadle soil volumetric moisture decreased by less than one percent. The Beadle soil under excess moisture allows that excess to drain away, therefore the moisture content of this soil stayed relatively constant throughout the two years of this study. This indicates that the Beadle soil denitrification levels should be equivalent among years that have typical or excess precipitate. The Worthing soil volumetric moisture content, on the other hand varies with the precipitation received. Denitrification was shown to increase rapidly with a precipitation event until the volumetric moisture content approaches saturated conditions (optimal for denitrification). Excess moisture quickly drains from the Beadle soil and it becomes too dry to denitrify.

A model for predicting denitrification is presented which is based upon the volumetric moisture content of the soils ( $\text{kg N ha}^{-1} \text{ day}^{-1} = -4.379 + 0.196 * \text{volumetric moisture}$ ). Volumetric moisture values are used in the form xx.x. The correlation coefficient (0.67) indicates a good fit for this model. The moisture value at which

denitrification ceases is 22.3%, derived by solving the equation for moisture with  $\text{Kg N ha}^{-1} \text{ day}^{-1}$  equal to zero. Thus when a soil is drier than 22.3% volumetric moisture, denitrification ceases. Even though this model is empirical, by using factors involved in the driving forces of denitrification, it is more mechanistic than many models.

The bulk density values of the soil sampled for the denitrification assay differed by soil type. Soil bulk density affects the amount of air filled spaces in the soil, and therefore affects the aeration of the soil. The Beadle soil bulk density averaged  $1.2 \text{ Mg m}^{-3}$  and the Worthing soil  $1.1 \text{ Mg m}^{-3}$ . The Beadle soil, a clay loam, contains more sand and less silt than does the Worthing soil. With a texture containing nearly equivalent amounts of each particle size, the Beadle soil compaction could result in a greater bulk density.

Denitrification on the Beadle soil was estimated at 13.8-kg nitrogen  $\text{ha}^{-1}$  evolved in 1986 and 30.25-kg in 1987. On the Worthing soil, estimated denitrification amounted to 22.25-kg nitrogen  $\text{ha}^{-1}$  evolved in 1986 and 28.5-kg in 1987. In dry years, the amount of nitrogen lost by denitrification would be equivalent on the two soils. In a wet year, considerably more nitrogen would be lost from the Worthing soil, since it has the poorer drainage and would be wet longer than the Beadle soil.

Tillage does play a part in the overall amount of nitrous oxide emission by the soil. Ridge tilled plots had higher nitrogen losses from denitrification than did the three other tillage plots (conventional, reduced, or no tillage), even though plant residue on the surface at the beginning of the season did not differ from that on the no till or reduced tillage plots. Within a Beadle and Worthing soil landscape, up to 40-kg nitrogen can be lost  $\text{ha}^{-1} \text{yr}^{-1}$ . An explanation of the higher rates on the ridge plots might be that the ridge acts like a wick, remaining moist longer and heating up faster than the soil under the other tillage methods. Another explanation might be that the ridge till plots had the majority of the residue present between the ridges, allowing the wet ridges to warm quickly. Conventional tillage and reduced tillage methods stir the soil more than the other two tillage methods. This mixes the soil, introducing air into the soil. The loss of aggregates and introduction of air would inhibit denitrification. One recommendation concerning farming this type of landscape would be to plow the soil thereby incorporating that residue, and to keep drainageways open so that the low lying soils could dry faster.

Surface 'glucose equivalent' amounts in these soils prior to and after cultivation were not different.

Breakdown of the abundant organic matter takes place at a

constant rate and the glucose (or glucose equivalent) appears to be readily available throughout the growing season.

Of the aggregate sizes studied, no one aggregate size denitrified more than another. Larger aggregates than those studied might denitrify more due to the larger anaerobic centers.

Adding 125- or 250-ug glucose  $\text{g}^{-1}$  soil had no effect on denitrification. These small glucose additions did not greatly increase the total soil glucose over that present previously. The addition of 625- and 1250-ug glucose  $\text{g}^{-1}$  produced highly significant differences over the zero control, increasing nitrous oxide emissions from 2790-ppm  $\text{g}^{-1}$  soil to 5100 to 7060 ppm. This data indicates that these glucose amendments stimulate an increased proliferation of the denitrifying organisms. Addition of more glucose than 1250 ug  $\text{g}^{-1}$  soil did not further increase nitrous oxide emissions. Higher glucose additions might have been greater than the organisms could use, or some other substance or factor became limiting.

Taking into account not only the relative cost of the fertilizer itself, along with the application costs of the machinery and labor, and the loss of yield potential, actual economic losses due to denitrification can be significant. When we add these to the environmental effects

of the increased nitrous oxide emissions, we realize that even though we do not know the net effect of raised levels of denitrification and what effect our tillage has on it, we need to be concerned about it from a soil management standpoint.

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Table 2. Denitrification rates in the Boreas well (June 2, 1987).

Fig. 1. Denitrification rates in the Boreas well.

Well	Location - T1			Depth (m)			Temperature (°C)			Denitrification Rate (mg N/m <sup>2</sup> /hr)		
	W1	W2	W3	W4	W5	W6	W7	W8	W9	W10	W11	W12
W1	111	112	113	114	115	116	117	118	119	120	121	122
W2	123	124	125	126	127	128	129	130	131	132	133	134
W3	135	136	137	138	139	140	141	142	143	144	145	146
W4	147	148	149	150	151	152	153	154	155	156	157	158
W5	159	160	161	162	163	164	165	166	167	168	169	170
W6	171	172	173	174	175	176	177	178	179	180	181	182
W7	183	184	185	186	187	188	189	190	191	192	193	194
W8	195	196	197	198	199	200	201	202	203	204	205	206
W9	207	208	209	210	211	212	213	214	215	216	217	218
W10	219	220	221	222	223	224	225	226	227	228	229	230
W11	231	232	233	234	235	236	237	238	239	240	241	242
W12	243	244	245	246	247	248	249	250	251	252	253	254
W13	255	256	257	258	259	260	261	262	263	264	265	266
W14	267	268	269	270	271	272	273	274	275	276	277	278
W15	279	280	281	282	283	284	285	286	287	288	289	290
W16	291	292	293	294	295	296	297	298	299	300	301	302
W17	303	304	305	306	307	308	309	310	311	312	313	314
W18	315	316	317	318	319	320	321	322	323	324	325	326
W19	327	328	329	330	331	332	333	334	335	336	337	338
W20	339	340	341	342	343	344	345	346	347	348	349	350
W21	351	352	353	354	355	356	357	358	359	360	361	362
W22	363	364	365	366	367	368	369	370	371	372	373	374
W23	375	376	377	378	379	380	381	382	383	384	385	386
W24	387	388	389	390	391	392	393	394	395	396	397	398
W25	399	400	401	402	403	404	405	406	407	408	409	410
W26	411	412	413	414	415	416	417	418	419	420	421	422
W27	423	424	425	426	427	428	429	430	431	432	433	434
W28	435	436	437	438	439	440	441	442	443	444	445	446
W29	447	448	449	450	451	452	453	454	455	456	457	458
W30	459	460	461	462	463	464	465	466	467	468	469	470
W31	471	472	473	474	475	476	477	478	479	480	481	482
W32	483	484	485	486	487	488	489	490	491	492	493	494
W33	495	496	497	498	499	500	501	502	503	504	505	506
W34	507	508	509	510	511	512	513	514	515	516	517	518
W35	519	520	521	522	523	524	525	526	527	528	529	530
W36	531	532	533	534	535	536	537	538	539	540	541	542
W37	543	544	545	546	547	548	549	550	551	552	553	554
W38	555	556	557	558	559	560	561	562	563	564	565	566
W39	567	568	569	570	571	572	573	574	575	576	577	578
W40	579	580	581	582	583	584	585	586	587	588	589	590
W41	591	592	593	594	595	596	597	598	599	600	601	602
W42	603	604	605	606	607	608	609	610	611	612	613	614
W43	615	616	617	618	619	620	621	622	623	624	625	626
W44	627	628	629	630	631	632	633	634	635	636	637	638
W45	639	640	641	642	643	644	645	646	647	648	649	650
W46	651	652	653	654	655	656	657	658	659	660	661	662
W47	663	664	665	666	667	668	669	670	671	672	673	674
W48	675	676	677	678	679	680	681	682	683	684	685	686
W49	687	688	689	690	691	692	693	694	695	696	697	698
W50	699	700	701	702	703	704	705	706	707	708	709	710
W51	711	712	713	714	715	716	717	718	719	720	721	722
W52	723	724	725	726	727	728	729	730	731	732	733	734
W53	735	736	737	738	739	740	741	742	743	744	745	746
W54	747	748	749	750	751	752	753	754	755	756	757	758
W55	759	760	761	762	763	764	765	766	767	768	769	770
W56	771	772	773	774	775	776	777	778	779	780	781	782
W57	783	784	785	786	787	788	789	790	791	792	793	794
W58	795	796	797	798	799	800	801	802	803	804	805	806
W59	807	808	809	810	811	812	813	814	815	816	817	818
W60	819	820	821	822	823	824	825	826	827	828	829	830
W61	831	832	833	834	835	836	837	838	839	840	841	842
W62	843	844	845	846	847	848	849	850	851	852	853	854
W63	855	856	857	858	859	860	861	862	863	864	865	866
W64	867	868	869	870	871	872	873	874	875	876	877	878
W65	879	880	881	882	883	884	885	886	887	888	889	890
W66	891	892	893	894	895	896	897	898	899	900	901	902
W67	903	904	905	906	907	908	909	910	911	912	913	914
W68	915	916	917	918	919	920	921	922	923	924	925	926
W69	927	928	929	930	931	932	933	934	935	936	937	938
W70	939	940	941	942	943	944	945	946	947	948	949	950
W71	951	952	953	954	955	956	957	958	959	960	961	962
W72	963	964	965	966	967	968	969	970	971	972	973	974
W73	975	976	977	978	979	980	981	982	983	984	985	986
W74	987	988	989	990	991	992	993	994	995	996	997	998
W75	999	1000	1001	1002	1003	1004	1005	1006	1007	1008	1009	1010

# APPENDIX A

## DENITRIFICATION

### RATE

Table 8. Denitrification rates in the Beadle soil (1986 & 1987).

Kg Nitrogen emitted at each sampling time

date	CONVENTIONAL TILL			REDUCED TILLAGE			RIDGE TILLAGE			NO TILLAGE		
	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3
86157	.030	.000	.005	.007	.009	.006	.450	.010	.018	.010	.008	.445
86165	.019	.005	.012	.142	.000	.170	.066	.223	.023	.052	.399	.253
86172	.015	.000	.001	.060	.000	.008	.198	.003	.148	.029	.012	.003
86183	.002	.005	.000	6.060	.066	2.847	.418	.000	.046	1.727	.136	3.526
86196	.001	.002	.001	.159	.009	.086	.031	.009	.035	.029	.002	.002
86206	.043	.076	.002	.223	.212	.002	.010	.222	.022	3.055	.188	.067
86213	.150	.009	.014	.018	.059	1.091	.062	.335	.104	4.250	.819	.244
86227	.282	3.593	.112	.058	1.954	2.722	1.026	4.979	8.065	.974	3.161	1.236
86314	.147	.032	2.267	.087	1.344	.027	.014	.173	.063	.392	.383	.044
AVE 86	.077	.414	.268	.757	.406	.773	.253	.662	.947	1.169	.567	.647
87159	.001	.001	.004	.003	.007	.007	.033	.058	.048	.002	.324	.023
87169	.031	.010	.003	.191	.138	.093	2.371	1.574	.291	1.226	1.585	.431
87178	1.586	1.285	.310	.360	.990	2.430	.970	2.310	1.340	2.010	1.510	3.540
87189	.003	.003	.001	.002	.464	.000	.858	.793	.038	.039	.015	.000
87203	.000	.047	.026	.003	.577	.072	.108	.700	.033	.015	.006	.046
87206	.181	3.780	3.381	.037	5.578	.703	2.793	2.896	2.651	3.385	2.864	5.822
87218	.012	1.159	1.760	3.974	.021	.465	1.903	.191	1.559	2.425	1.249	1.270
87220	.024	1.788	.043	3.292	4.319	1.111	2.882	1.386	.909	3.437	.327	.123
87225	1.241	2.136	1.409	3.648	2.962	1.145	1.929	1.361	1.392	1.940	1.408	1.175
87232	.907	.650	.016	1.538	1.355	.111	.011	1.172	1.185	1.513	1.640	.134
87253	2.577	1.755	1.734	1.791	2.624	1.715	1.652	2.200	1.855	1.636	2.420	.352
87267	.003	.001	.010	.001	.001	.002	8.480	.002	.001	.008	.003	.003
87281	1.064	1.000	.646	.064	.001	.478	1.968	.549	.002	1.301	.097	2.936
87290	.067	.137	.681	.926	.343	1.790	2.068	5.174	2.446	.830	2.539	1.093
AVE 87	.550	.982	.716	1.131	1.384	.723	2.002	1.455	.982	1.412	1.142	1.211
AVERAGE	.353	.745	.529	.975	.977	.744	1.273	1.124	.968	1.311	.903	.976



Table 9. Denitrification rates in the Worthing soil (1986 & 1987).

Kg Nitrogen emitted at each sampling time

date	CONVENTIONAL TILL			REDUCED TILLAGE			RIDGE TILLAGE			NO TILLAGE		
	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3
	151	251	351	161	261	361	171	271	371	181	281	381
86157	.670	.291	.069	.695	.505	.342	.095	.023	.334	.320	.904	.072
86165	.683	.306	.070	.730	.557	.384	.099	.023	.343	.336	.956	.076
86172	.230	.290	.381	.056	.098	.113	.692	.153	4.744	.662	.354	.790
86183	1.699	.114	.292	3.844	3.212	3.893	5.930	1.887	1.859	.395	4.923	3.895
86196	.031	.032	.092	.083	.087	.081	.002	.166	.002	.087	.795	1.031
86206	4.266	.925	.813	3.903	.802	.256	4.313	.412	6.185	.439	.416	4.155
86213	.422	.410	1.646	.558	1.260	1.793	3.163	2.908	10.639	.857	.264	.275
86227	.935	.514	.380	.460	3.265	.233	4.478	4.069	1.762	.030	.115	.224
86314	.197	.164	.131	.165	.268	.247	.112	.819	.187	.336	.218	.296
AVE 86	1.015	.338	.430	1.166	1.117	.816	2.098	1.162	2.895	.385	.994	1.202
87159	.056	.342	.009	.003	.004	.003	1.173	.000	.122	.001	.015	.154
87169	.098	.227	.303	.858	.423	1.436	2.539	.317	.628	.494	.078	.256
87178	.940	1.380	.070	1.620	.460	1.050	4.600	.420	.600	1.250	3.610	.680
87189	.004	.014	.004	.017	.275	.009	.000	.003	.466	.005	.000	.505
87203	.003	.005	.006	.036	.016	.012	1.436	.035	.864	.000	.236	1.504
87206	.334	1.491	2.388	.354	.335	.418	2.994	.138	1.119	.307	2.129	2.554
87218	4.088	4.908	1.317	.371	7.923	7.270	8.587	.615	3.766	1.502	.426	.926
87220	.627	3.343	.014	.907	2.444	.648	1.500	2.468	.530	.147	4.069	.297
87225	2.843	2.829	1.883	2.729	1.000	2.041	7.832	.274	1.564	.165	.578	.173
87232	.605	1.798	.792	2.744	.473	1.220	2.535	1.311	.256	.812	.503	.113
87253	1.601	1.512	1.480	2.705	1.596	.698	3.439	.973	2.822	.368	.427	.191
87267	.000	.003	.004	.002	.013	.000	.019	.004	.003	.004	.331	.081
87281	.014	.007	.484	.110	.966	.651	.527	.000	1.596	.054	.047	.607
87290	1.062	1.009	.217	2.579	.380	.200	3.608	.064	1.630	.218	.224	.244
AVE 87	.877	1.348	.641	1.074	1.165	1.118	2.914	.473	1.140	.381	.905	.592
AVERAGE	.934	.927	.553	1.112	1.145	.992	2.574	.760	1.872	.382	.942	.846

Table 10. Volumetric moisture values in the Beadle soil (1986 & 1987).

Volumetric moisture of each sample

date	CONVENTIONAL TILL			REDUCED TILLAGE			RIDGE TILLAGE			NO TILLAGE		
	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3
86157	29.03	24.24	26.44	28.68	30.60	25.84	32.10	27.47	30.14	24.59	30.24	31.97
86165	32.72	27.05	-----	33.23	32.60	25.88	37.56	32.84	30.21	31.53	28.47	32.17
86172	27.06	20.15	23.46	22.39	22.14	26.35	24.13	21.76	30.37	24.86	27.27	27.37
86183	24.42	24.05	28.02	30.88	28.91	24.57	24.41	20.66	30.59	31.36	23.14	30.83
86196	22.73	20.83	25.08	30.13	26.40	23.42	27.90	20.83	26.01	23.50	30.71	21.82
86206	24.98	27.33	29.25	33.45	27.69	29.82	22.43	28.85	27.05	28.87	28.14	32.17
86213	39.55	38.80	38.49	39.38	39.45	41.33	38.23	39.61	39.75	42.52	38.17	38.29
86227	31.86	42.44	37.00	33.97	40.22	31.88	31.29	33.11	40.65	38.41	33.79	40.50
86314	34.53	41.80	41.00	36.51	39.99	36.67	25.79	39.23	37.89	36.92	40.58	33.56
AVE 86	29.65	29.63	31.09	32.07	32.00	29.53	29.32	29.37	32.52	31.40	31.17	32.06
87159	19.05	17.50	18.43	23.23	19.80	23.52	23.67	24.57	26.92	21.36	23.58	22.11
87169	36.35	31.69	34.52	37.80	38.88	40.44	37.20	43.21	42.14	39.38	41.25	44.43
87178	38.82	41.39	44.08	38.82	41.84	43.31	35.72	39.47	44.53	34.93	43.42	44.12
87189	14.63	15.19	16.35	16.41	15.09	14.28	17.67	19.92	17.89	20.58	18.17	14.77
87203	21.64	18.60	18.29	12.82	27.09	20.75	22.29	20.56	14.26	19.79	16.41	22.67
87206	31.50	35.78	35.55	30.11	39.40	38.68	37.89	35.80	37.34	36.28	32.84	39.81
87218	33.04	33.61	36.87	33.33	33.21	38.37	31.06	32.46	33.87	30.81	38.11	39.83
87220	34.16	34.38	34.06	35.53	36.20	37.52	34.18	34.68	35.09	37.60	37.46	39.36
87225	34.93	40.03	42.30	37.74	38.88	41.31	37.70	41.09	43.66	40.82	42.18	40.42
87232	19.02	24.43	23.83	22.85	30.23	22.91	17.20	17.61	21.05	22.48	20.91	33.81
87253	26.68	30.02	35.39	28.89	32.27	35.07	24.86	25.93	27.86	33.45	25.83	32.72
87267	11.53	18.48	24.23	13.82	17.00	17.02	17.95	11.95	15.90	17.52	18.92	22.81
87281	27.27	29.25	32.27	27.13	22.51	24.82	23.99	17.48	27.07	29.07	28.97	32.54
87290	36.95	43.52	43.19	40.05	41.31	42.61	33.00	39.97	41.23	42.65	39.45	41.73
AVE 87	27.54	29.56	31.38	28.47	30.98	31.47	28.17	28.91	30.63	30.48	30.54	33.65
AVERAGE	28.42	29.59	31.27	29.97	31.40	30.66	28.65	29.10	31.42	30.86	30.80	32.99

Table 11. Volumetric moisture values in the Worthing soil (1986 & 1987).

Volumetric moisture of each sample

date	CONVENTIONAL TILL			REDUCED TILLAGE			RIDGE TILLAGE			NO TILLAGE		
	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3
86157	49.40	51.09	37.04	48.91	48.41	48.21	38.90	42.76	47.75	46.01	43.96	48.65
86165	51.25	52.37	50.17	47.98	49.59	46.35	48.22	51.46	48.65	50.43	47.04	48.59
86172	39.87	41.44	34.87	37.83	45.28	38.76	33.24	41.27	46.31	40.53	38.91	35.13
86183	46.78	46.25	39.67	39.93	39.53	41.65	39.91	37.24	45.79	40.07	40.44	39.99
86196	48.82	48.89	33.23	41.98	37.11	39.53	41.45	37.48	31.64	34.40	38.01	34.77
86206	51.80	53.24	50.73	46.11	43.19	46.86	50.24	47.31	49.59	45.91	42.40	47.31
86213	51.50	46.45	54.03	51.62	53.95	52.51	51.38	51.18	51.80	49.92	47.39	48.78
86227	53.36	52.79	50.46	51.14	53.28	51.17	42.41	51.19	47.55	46.73	44.55	43.27
86314	52.61	52.15	51.69	55.92	52.05	51.39	52.28	50.25	52.05	52.29	51.99	51.80
AVE 86	49.49	49.41	44.65	46.82	46.93	46.27	44.23	45.57	46.79	45.14	43.85	44.25
87159	28.64	29.42	29.04	18.26	23.43	29.44	39.52	29.37	37.52	26.14	30.89	40.42
87169	39.87	42.63	38.31	50.41	51.76	46.08	48.32	46.98	50.16	50.06	41.73	46.56
87178	49.13	42.87	44.63	52.13	50.77	51.17	49.43	49.03	47.67	46.52	49.92	47.29
87189	20.54	24.17	7.74	23.76	26.88	20.12	36.69	24.94	33.79	26.18	25.06	33.41
87203	23.68	19.17	25.32	28.99	27.43	20.34	29.72	23.22	35.01	23.30	23.58	31.04
87206	43.86	40.66	45.34	43.36	42.93	43.56	40.03	47.10	42.95	41.94	46.70	43.50
87218	39.34	40.50	43.36	43.21	49.13	41.39	43.52	43.42	39.53	42.58	43.35	45.58
87220	43.07	39.44	41.19	45.50	46.21	44.33	45.68	44.17	43.01	43.42	44.43	42.42
87225	55.27	52.53	56.83	55.33	49.57	56.22	52.39	53.57	44.96	52.31	55.27	46.39
87232	37.66	38.07	32.96	34.04	35.70	43.88	31.26	37.76	31.83	24.28	28.09	36.87
87253	30.17	37.07	27.55	33.29	40.68	39.44	36.93	31.48	33.29	37.89	40.90	39.34
87267	20.66	18.42	21.27	18.19	15.34	21.23	23.22	21.35	20.02	20.81	25.83	32.46
87281	33.95	32.44	30.81	35.94	37.72	33.61	33.73	29.34	29.96	36.14	36.14	39.18
87290	44.81	41.07	44.91	49.78	50.91	46.52	45.18	46.33	41.63	45.77	46.48	46.36
AVE 87	36.48	35.60	34.95	38.01	39.18	38.38	39.69	37.72	37.95	36.95	38.46	40.77
AVERAGE	41.90	41.36	38.99	41.68	42.41	41.67	41.58	40.99	41.64	40.37	40.70	42.22



Table 12. Bulk density values in the Beadle soil (1986 &amp; 1987).

date	CONVENTIONAL TILLAGE			REDUCED TILLAGE			RIDGE TILLAGE			NO TILLAGE		
	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3
	(Mg m <sup>-3</sup> )											
86157	1.28	1.09	1.13	1.26	1.22	1.16	1.47	1.24	1.08	1.19	1.12	1.21
86165	1.26	1.05	----	1.18	1.27	1.14	1.29	1.24	1.15	1.16	1.03	1.11
86172	1.17	.98	1.09	1.12	1.07	1.14	1.33	1.29	1.13	1.15	1.21	1.20
86183	1.27	1.09	1.04	1.22	1.15	1.07	1.25	1.03	1.21	1.26	1.07	1.19
86196	1.14	1.04	1.06	1.22	1.08	1.09	1.30	1.10	1.06	1.07	1.21	1.09
86206	1.25	1.09	1.18	1.20	1.11	1.23	1.18	1.21	1.18	1.13	1.27	1.29
86213	1.27	1.26	1.26	1.27	1.26	1.25	1.32	1.33	1.22	1.28	1.23	1.27
86227	1.30	1.25	1.12	1.27	1.23	1.24	1.22	1.21	1.17	1.21	1.26	1.24
86314	1.32	1.28	1.22	1.32	1.18	1.24	1.26	1.30	1.22	1.27	1.22	1.12
AVE 86	1.27	1.13	1.15	1.26	1.18	1.19	1.29	1.24	1.19	1.21	1.20	1.22
87158	1.12	1.08	1.09	1.04	1.11	1.21	1.23	1.22	1.16	1.19	1.20	1.12
87169	1.30	1.16	1.19	1.36	1.21	1.25	1.25	1.30	1.22	1.15	1.21	1.31
87178	1.34	1.19	1.09	1.29	1.30	1.29	1.30	1.18	1.32	1.29	1.26	1.41
87189	1.14	1.05	1.05	1.09	1.00	1.16	1.19	1.08	.92	1.14	1.22	1.14
87203	1.22	1.03	1.04	1.06	1.13	1.05	1.28	1.23	.91	1.25	1.09	1.13
87206	1.31	1.30	1.30	1.13	1.20	1.19	1.33	1.08	1.25	1.21	1.23	1.28
87218	1.31	1.16	1.26	1.33	1.14	1.28	1.24	1.24	1.22	1.12	1.24	1.34
87220	1.34	1.17	1.16	1.36	1.24	1.23	1.36	1.25	1.16	1.23	1.25	1.31
87225	1.39	1.23	1.26	1.33	1.25	1.25	1.38	1.30	1.24	1.29	1.23	1.21
87232	1.07	1.10	.98	1.24	1.20	1.04	1.18	1.06	1.01	1.05	.93	1.24
87253	1.25	1.23	1.24	1.24	1.24	1.27	1.13	1.18	1.14	1.05	1.13	1.31
87267	1.13	1.06	1.05	1.02	1.08	1.04	1.06	1.08	.97	1.15	1.05	1.19
87281	1.24	1.11	1.13	1.24	1.16	1.03	1.10	1.04	1.03	1.17	1.14	1.30
87290	.07	1.35	1.35	1.24	1.36	1.32	1.28	1.25	1.37	1.29	1.26	1.29
AVE 87	1.16	1.16	1.16	1.21	1.19	1.19	1.24	1.18	1.14	1.18	1.17	1.26
AVERAGE	1.21	1.15	1.15	1.23	1.18	1.19	1.26	1.21	1.16	1.20	1.19	1.24

Table 13. Bulk density values in the Worthing soil (1986 &amp; 1987).

(Mg m <sup>-3</sup> )												
date	CONVENTIONAL TILL			REDUCED TILLAGE			RIDGE TILLAGE			NO TILLAGE		
	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3	REP1	REP2	REP3
86157	1.12	1.08	1.13	1.06	1.16	1.14	1.20	1.19	1.19	1.19	1.12	1.30
86165	1.06	.96	.98	1.06	.94	.90	1.03	1.09	1.12	1.05	.97	1.18
86172	1.05	1.05	.99	1.02	1.10	1.19	1.01	1.15	1.17	1.17	1.14	1.01
86183	1.12	1.09	1.00	1.01	1.02	1.01	1.14	.97	1.11	1.13	1.15	1.12
86196	1.11	1.07	.85	1.03	.97	1.09	1.21	1.07	1.12	.99	1.15	1.04
86206	1.10	1.09	1.07	1.10	1.06	1.09	1.13	1.16	1.20	1.17	1.15	1.12
86213	1.03	1.01	1.13	1.14	1.07	1.12	1.18	1.20	1.17	1.20	1.21	1.12
86227	1.11	1.17	1.12	1.02	1.11	1.12	1.19	1.15	1.21	1.21	1.23	1.16
86314	1.08----		1.05	1.06	1.03	1.05	1.12	1.14	1.11	1.12	1.17	1.15
AVE 86	1.09	1.07	1.04	1.06	1.05	1.08	1.13	1.12	1.16	1.14	1.14	1.13
87159	.96	.96	1.03	.91	.98	1.19	1.07	1.12	1.12	1.08	1.03	1.08
87169	1.12	1.11	1.08	1.10	1.15	1.16	1.05	1.12	1.15	1.12	.96	1.18
87178	1.25	1.18	1.20	1.14	1.01	1.23	1.11	1.18	1.24	1.20	1.25	1.25
87189	.96	1.04	.81	.93	.99	1.01	1.15	1.09	1.11	1.07	1.11	1.19
87203	1.00	.97	.89	.98	1.01	1.02	.96	1.08	1.18	1.34	.99	1.15
87206	1.20	1.08	1.11	1.24	1.20	1.20	1.18	1.17	1.15	1.13	1.22	1.21
87218	1.12	1.21	1.16	1.10	1.17	1.19	1.18	1.27	1.20	1.27	1.26	1.18
87220	1.18	1.14	1.11	1.16	1.10	1.20	1.25	1.26	1.21	1.29	1.26	1.21
87225	1.11	1.10	1.09	1.02	.91	1.13	1.10	1.06	1.14	1.09	1.02	1.23
87232	1.13	1.16	1.01	1.19	1.04	1.15	1.11	1.09	1.02	1.24	1.22	1.14
87253	1.13	1.15	1.07	1.04	1.11	1.17	1.18	1.08	1.06	1.17	1.19	1.25
87267	.99	1.00	.97	.90	1.08	1.09	1.01	1.02	1.06	.99	1.08	1.21
87281	1.12	1.06	1.01	1.08	1.03	1.07	1.07	1.05	1.03	1.16	1.16	1.32
87290	1.18	1.07	1.13	1.20	1.19	1.25	1.07	1.21	1.10	1.25	1.25	1.16
AVE 87	1.10	1.09	1.05	1.07	1.07	1.15	1.11	1.13	1.13	1.17	1.14	1.20
AVERAGE	1.10	1.08	1.04	1.06	1.06	1.12	1.12	1.13	1.14	1.16	1.14	1.17



Table 15. Denitrification by mixed aggregate size.

Run	Denitrification rate (g N/g dry wt. d)	Aggregate size (mm)	Denitrification rate (g N/g dry wt. d)	Aggregate size (mm)	Denitrification rate (g N/g dry wt. d)	Aggregate size (mm)
<b>Denitrification by mixed aggregate size</b>						
Run 1	1.0	0.5	1.0	0.5	1.0	0.5
Run 2	1.0	0.5	1.0	0.5	1.0	0.5
Run 3	1.0	0.5	1.0	0.5	1.0	0.5
Run 4	1.0	0.5	1.0	0.5	1.0	0.5
Run 5	1.0	0.5	1.0	0.5	1.0	0.5
Run 6	1.0	0.5	1.0	0.5	1.0	0.5
Run 7	1.0	0.5	1.0	0.5	1.0	0.5
Run 8	1.0	0.5	1.0	0.5	1.0	0.5
Run 9	1.0	0.5	1.0	0.5	1.0	0.5
Run 10	1.0	0.5	1.0	0.5	1.0	0.5
Run 11	1.0	0.5	1.0	0.5	1.0	0.5
Run 12	1.0	0.5	1.0	0.5	1.0	0.5
Run 13	1.0	0.5	1.0	0.5	1.0	0.5
Run 14	1.0	0.5	1.0	0.5	1.0	0.5
Run 15	1.0	0.5	1.0	0.5	1.0	0.5
Run 16	1.0	0.5	1.0	0.5	1.0	0.5
Run 17	1.0	0.5	1.0	0.5	1.0	0.5
Run 18	1.0	0.5	1.0	0.5	1.0	0.5
Run 19	1.0	0.5	1.0	0.5	1.0	0.5
Run 20	1.0	0.5	1.0	0.5	1.0	0.5
Run 21	1.0	0.5	1.0	0.5	1.0	0.5
Run 22	1.0	0.5	1.0	0.5	1.0	0.5
Run 23	1.0	0.5	1.0	0.5	1.0	0.5
Run 24	1.0	0.5	1.0	0.5	1.0	0.5
Run 25	1.0	0.5	1.0	0.5	1.0	0.5
Run 26	1.0	0.5	1.0	0.5	1.0	0.5
Run 27	1.0	0.5	1.0	0.5	1.0	0.5
Run 28	1.0	0.5	1.0	0.5	1.0	0.5
Run 29	1.0	0.5	1.0	0.5	1.0	0.5
Run 30	1.0	0.5	1.0	0.5	1.0	0.5
Run 31	1.0	0.5	1.0	0.5	1.0	0.5
Run 32	1.0	0.5	1.0	0.5	1.0	0.5
Run 33	1.0	0.5	1.0	0.5	1.0	0.5
Run 34	1.0	0.5	1.0	0.5	1.0	0.5
Run 35	1.0	0.5	1.0	0.5	1.0	0.5
Run 36	1.0	0.5	1.0	0.5	1.0	0.5
Run 37	1.0	0.5	1.0	0.5	1.0	0.5
Run 38	1.0	0.5	1.0	0.5	1.0	0.5
Run 39	1.0	0.5	1.0	0.5	1.0	0.5
Run 40	1.0	0.5	1.0	0.5	1.0	0.5
Run 41	1.0	0.5	1.0	0.5	1.0	0.5
Run 42	1.0	0.5	1.0	0.5	1.0	0.5
Run 43	1.0	0.5	1.0	0.5	1.0	0.5
Run 44	1.0	0.5	1.0	0.5	1.0	0.5
Run 45	1.0	0.5	1.0	0.5	1.0	0.5
Run 46	1.0	0.5	1.0	0.5	1.0	0.5
Run 47	1.0	0.5	1.0	0.5	1.0	0.5
Run 48	1.0	0.5	1.0	0.5	1.0	0.5
Run 49	1.0	0.5	1.0	0.5	1.0	0.5
Run 50	1.0	0.5	1.0	0.5	1.0	0.5

APPENDIX D  
DENITRIFICATION BY  
AGGREGATE SIZE

Table 14. Nitrous oxide production by sieved aggregate size.

SITE	Aggregate size (mm)	mg N <sub>2</sub> /Kg soil REP 1	mg N <sub>2</sub> /Kg soil REP 2	SITE	Aggregate size (mm)	mg N <sub>2</sub> /Kg soil REP 1	mg N <sub>2</sub> /Kg soil REP 2
CONVENTIONAL TILLAGE				RIDGE TILLAGE			
WORTHING 4	<1	.002	2.550	WORTHING 1	<1	.0291	.8468
	1 TO 2	.065	.096		1 TO 2	0	.0933
	2 TO 4.75	.016	.018		2 TO 4.75	.1586	.1337
	4.75 TO 6.36	.000	1.205		4.75 TO 6.36	.0544	.5596
	6.36 TO 9.53	.000	.304		6.36 TO 9.53	0	.0707
WORTHING 21	<1	.053	.977	WORTHING 16	<1	0	.1449
	1 TO 2	.751	.238		1 TO 2	3.481	2.060
	2 TO 4.75	.043	.191		2 TO 4.75	.4133	.6084
	4.75 TO 6.36	.075	.000		4.75 TO 6.36	.1354	.4034
	6.36 TO 9.53	.085	.764		6.36 TO 9.53	0	.1439
WORTHING 25	<1	.015	.281	WORTHING 34	<1	.0806	4.928
	1 TO 2	.237	1.633		1 TO 2	1.202	3.905
	2 TO 4.75	.015	.656		2 TO 4.75	.0775	6.507
	4.75 TO 6.36	.044	.646		4.75 TO 6.36	.0738	3.397
	6.36 TO 9.53	.021	1.935		6.36 TO 9.53	0	2.891
REDUCED TILLAGE				NO TILLAGE			
WORTHING 8	<1	.024	.709	WORTHING 12	<1	.0050	2.660
	1 TO 2	.093	.128		1 TO 2	.3138	.0938
	2 TO 4.75	.008	4.294		2 TO 4.75	.2639	.4703
	4.75 TO 6.36	.032	1.035		4.75 TO 6.36	1.267	0
	6.36 TO 9.53	.044	.494		6.36 TO 9.53	.0282	.5320
WORTHING 24	<1	.079	.322	WORTHING 14	<1	.0828	1.753
	1 TO 2	.199	.115		1 TO 2	.0379	1.131
	2 TO 4.75	.016	3.469		2 TO 4.75	0	2.073
	4.75 TO 6.36	.000	.116		4.75 TO 6.36	.0644	.0178
	6.36 TO 9.53	.000	1.704		6.36 TO 9.53	1.545	.1900
WORTHING 28	<1	.334	.803	WORTHING 31	<1	0	1.880
	1 TO 2	.148	.201		1 TO 2	0	2.986
	2 TO 4.75	.783	.089		2 TO 4.75	.0679	2.202
	4.75 TO 6.36	1.378	6.577		4.75 TO 6.36	.1342	.2203
	6.36 TO 9.53	.024	7.593		6.36 TO 9.53	.1131	.1599

Table 15. Average temperatures of incubation for each sampling date.

DATE	TEMPERATURE (°C)
07/10/80	13
07/11/80	14
07/12/80	15
07/13/80	16
07/14/80	17
07/15/80	18
07/16/80	19
07/17/80	20
07/18/80	21
07/19/80	22
07/20/80	23
07/21/80	24
07/22/80	25
07/23/80	26
07/24/80	27
07/25/80	28
07/26/80	29
07/27/80	30
07/28/80	31
07/29/80	32
07/30/80	33
07/31/80	34
08/01/80	35
08/02/80	36
08/03/80	37
08/04/80	38
08/05/80	39
08/06/80	40
08/07/80	41
08/08/80	42
08/09/80	43
08/10/80	44
08/11/80	45
08/12/80	46
08/13/80	47
08/14/80	48
08/15/80	49
08/16/80	50
08/17/80	51
08/18/80	52
08/19/80	53
08/20/80	54
08/21/80	55
08/22/80	56
08/23/80	57
08/24/80	58
08/25/80	59
08/26/80	60
08/27/80	61
08/28/80	62
08/29/80	63
08/30/80	64
08/31/80	65

# APPENDIX E

## TEMPERATURES OF INCUBATION

Table 15. Average temperatures of incubation for each sampling date.

DATE	TEMPERATURE (°C)
5/ 7/86	9
5/14/86	13
5/21/86	14
6/ 2/86	15
6/15/86	20
6/25/86	18
7/ 1/86	15
7/15/86	21
10/16/86	5
5/ 7/87	17
5/18/87	17
5/27/87	17
6/ 8/87	20
6/22/87	20
6/25/87	20
7/ 6/87	21.5
7/ 8/87	21.5
7/13/87	21.5
7/20/87	21.5
8/10/87	21
8/24/87	21
9/ 9/87	20
9/17/87	20

Table 18. *Analysis of variance for the effect of treatment on the yield of wheat (Triticum aestivum L.) in the 1954-55 season. The experiment was conducted at the Agricultural Research Station, Ludhiana, Punjab, India. The treatments were: (1) Control, (2) Fertilizer, (3) Fertilizer + Pesticide, (4) Fertilizer + Pesticide + Insecticide, (5) Fertilizer + Pesticide + Insecticide + Fungicide. The results are given in Table 18.*

Analysis of variance for the effect of treatment on the yield of wheat (Triticum aestivum L.) in the 1954-55 season				
Treatment	Yield (kg/ha)	Standard Error	D.F.	F-value
Control	1.45	0.045	10	0.000
Fertilizer	1.85	0.045	10	0.000
Fertilizer + Pesticide	1.95	0.045	10	0.000
Fertilizer + Pesticide + Insecticide	2.05	0.045	10	0.000
Fertilizer + Pesticide + Insecticide + Fungicide	2.15	0.045	10	0.000

## APPENDIX F

## ANOVA TABLES

Table 16. ANOVA for the evaluation of the effect of soils (Beadle and Worthing) and tillage (conventional, ridge till, reduced till, and no till) on denitrification rate. Three reps were used on each soil.

## ANOVA

Dependent Variable = denitrification rate

<u>SOURCE</u>	<u>D.F.</u>	<u>Mean Square</u>	<u>F</u>	<u>PR &gt; F</u>
Tillage	3	0.5940	2.63	0.2245
Soil	1	0.1610	2.36	0.1989
Tillage * Soil	3	0.2263	1.35	0.3052
Rep (Soil)	4	0.0681	0.41	0.8010
Error	12	0.1678		



Table 17. ANOVA for the evaluation of the effect of soils (Beadle and Worthing) and tillage (conventional, ridge till, reduced till, and no till) on volumetric moisture. Three reps were used on each soil.

ANOVA				
Dependent Variable = volumetric moisture				
<u>SOURCE</u>	<u>D.F.</u>	<u>Mean Square</u>	<u>F</u>	<u>PR &gt; F</u>
Tillage	3	1.9152	1.54	0.3648
Soil	1	683.3068	279.43	0.0001
Tillage * Soil	3	1.2401	1.41	0.2868
Rep (Soil)	4	2.4454	2.79	0.0754
Error	12	.8770		

Table 18. ANOVA for the evaluation of the effect of soils (Beadle and Worthing) and tillage (conventional, ridge till, reduced till, and no till) on bulk density. Three reps were used on each soil.

ANOVA

Dependent Variable = bulk density

<u>SOURCE</u>	<u>D.F.</u>	<u>Mean Square</u>	<u>F</u>	<u>PR &gt; F</u>
Tillage	3	.0025	1.11	0.4667
Soil	1	.0376	16.68	0.0150
Tillage * Soil	3	.0022	1.49	0.2673
Rep (Soil)	4	.0023	1.52	0.2592
Error	12	.0015		

Table 19. ANOVA for the evaluation of the effect of soils (Beadle and Worthing), tillage (conventional, ridge till, reduced till, and no till), and year (1986 and 1987) on bulk density. Three reps were used on each soil.

## ANOVA

Dependent Variable = Bulk Density

<u>SOURCE</u>	<u>D.F.</u>	<u>Mean Square</u>	<u>F</u>	<u>PR &gt; F</u>
TREATMENT	3	.00157	2.38	0.1309
SOIL	1	.00016	0.24	0.6374
YEAR	1	.02806	42.43	0.0001
ERROR	10	.00066		

Table 20. ANOVA for the evaluation of the effect of soils (Beadle and Worthing), tillage (conventional, ridge till, reduced till, and no till), and year (1986 and 1987) on pH. Three reps were used on each soil.

## ANOVA

Dependent Variable = pH

<u>SOURCE</u>	<u>D.F.</u>	<u>Mean Square</u>	<u>F</u>	<u>PR &gt; F</u>
TREATMENT	3	0.2542	2.92	0.0866
SOIL	1	1.5625	17.96	0.0017
YEAR	1	0.2025	2.33	0.1581
ERROR	10	0.0870		

Table 21. ANOVA for the evaluation of the effect of soils (Beadle and Worthing), tillage (conventional, ridge till, reduced till, and no till), and year (1986 and 1987) on residue. Three reps were used on each soil.

ANOVA					
Dependent Variable = Residue					
SOURCE	D.F.	Mean Square	F	PR > F	
TREATMENT	3	1214.0833	6.52	0.0101	
SOIL	1	1764.0000	9.48	0.0117	
YEAR	1	342.2500	1.84	0.2049	
ERROR	10	186.1250			

Table 22. ANOVA for the evaluation of the effect of soils (Beadle and Worthing), tillage (conventional, ridge till, reduced till, and no till), and year (1986 and 1987) on residue cover. Three reps were used on each soil.

ANOVA				
Dependent Variable = Residue				
<u>SOURCE</u>	<u>D.F.</u>	<u>Mean Square</u>	<u>F</u>	<u>PR &gt; F</u>
TILLAGE	3	1214.00	9.99	0.0453
SOIL	1	1764.00	14.52	0.0318
YEAR	1	342.00	2.82	0.1919
TILLAGE * SOIL	3	232.80	1.92	0.3033
TILLAGE * YEAR	3	89.75	0.74	0.5953
SOIL * YEAR	1	529.00	4.35	0.1282
TILLAGE * SOIL *YEAR	3	121.50		



Table 23. ANOVA for the evaluation of the effect of soils (Beadle and Worthing), tillage (conventional, ridge till, reduced till, and no till), and year (1986 and 1987) on bulk density. Three reps were used on each soil.

ANOVA				
Dependent Variable = Bulk Density				
<u>SOURCE</u>	<u>D.F.</u>	<u>Mean Square</u>	<u>F</u>	<u>PR &gt; F</u>
TILLAGE	3	.00157	27.96	0.0108
SOIL	1	.00015	2.78	0.1942
YEAR	1	.02806	498.78	0.0002
TILLAGE * SOIL	3	.00052	9.00	0.0520
TILLAGE * YEAR	3	.00147	26.19	0.0118
SOIL * YEAR	1	.00051	9.00	0.0577
TILLAGE * SOIL * YEAR	3	.000056		

Table 24. Covariant ANOVA for the evaluation of the effect of soils (Beadle and Worthing), tillage (conventional, ridge till, reduced till, and no till), and volumetric moisture on denitrification rate. Three reps were used on each soil.

COVARIANT ANOVA				
Dependent variable = Denitrification rate				
Covariant = Volumetric moisture				
<u>SOURCE</u>	<u>D.F.</u>	<u>Mean Square</u>	<u>F</u>	<u>PR &gt; F</u>
TILLAGE	3	.6258	10.31	0.0434
SOIL	1	.3210	2.19	0.2131
TILLAGE * SOIL	3	.0607	0.41	0.7491
REP (SOIL)	4	.1466	0.99	0.4526
VOL. MOISTURE	1	.3853	2.60	0.1350
ERROR	11	.1481		

Table 25. Covariant ANOVA for the evaluation of the effect of soils (Beadle and Worthing), tillage (conventional, ridge till, reduced till, and no till), volumetric moisture, and bulk density on denitrification rate. Three reps were used on each soil.

## COVARIANT ANOVA

Dependent variable = Denitrification rate

Covariants = Volumetric moisture and Bulk density

<u>SOURCE</u>	<u>D.F.</u>	<u>Mean Square</u>	<u>F</u>	<u>PR &gt; F</u>
TILLAGE	3	.6452	11.26	0.0386
SOIL	1	.3525	2.33	0.2012
TILLAGE * SOIL	3	.0573	0.36	0.7865
REP (SOIL)	4	.1513	0.95	0.4727
VOL. MOISTURE	1	.4014	2.52	0.1432
BULK DENSITY	1	.0350	0.22	0.6466
ERROR	10	.1595		

Table 27. ANOVA for the evaluation of the effect of tillage (conventional, reduced till, ridge till, and no till), and time (pre- and post-cultivation) on glucose production. Three reps were used on each soil.

Table 26. ANOVA for the evaluation of the effect of soils (Beadle and Worthing), tillage (conventional, ridge till, reduced till, and no till), and time (pre- and post-cultivation) on glucose production. Three reps were used on each soil.

ANOVA				
Dependent Variable = Pre- and Post-Cultivation Glucose				
SOURCE	D.F.	Mean Square	F	PR > F
SOIL	1	24003.4	1.85	0.2459
TILLAGE	3	7856.8	0.55	0.6825
SOIL*TILLAGE	3	14298.9	2.33	0.1255
TIME	1	2.1	0.00	0.9844
SOIL*TIME	1	7280.2	1.46	0.2666
TILLAGE*TIME	3	2738.0	0.55	0.6652
SOIL*TILLAGE*TIME	3	6097.3	1.22	0.3711
REP(SOIL)	4	13009.9	2.60	0.1273
TILLAGE*REP(SOIL)	12	6123.8	1.23	0.4083
TIME*REP(SOIL)	4	1440.1	0.29	0.8767
ERROR	7	4995.9		

Table 27. MANOVA for the evaluation of the effect of tillage (conventional, reduced till, ridge till, and no till), and aggregate size (0 to 1mm, 1 to 2mm, 2 to 4.75mm, 4.75 to 6.36mm, and 6.36 to 9.53mm) on nitrous oxide production. Two reps were used on each aggregate size. Only the Worthing soil was used.

## ANOVA

Dependent Variable = Nitrous Oxide Production

for Five Aggregate Sizes

SIZE & SOURCE	D.F.	Mean Square	F	PR > F
<u>0 to 1 mm</u>				
TILLAGE	3	.0070	.17	0.9142
REP	1	.3725	9.06	0.0083
TILLAGE * REP	3	.0127	.31	0.8185
ERROR	16	.0411		
<u>1 to 2 mm</u>				
TILLAGE	3	.7761	1.95	0.1620
REP	1	.5116	1.29	0.2734
TILLAGE * REP	3	.4747	1.19	0.3437
ERROR	16	.3976		
<u>2 to 4.75 mm</u>				
TILLAGE	3	.1055	1.06	0.3939
REP	1	.1770	1.78	0.2013
TILLAGE * REP	3	.0173	.17	0.9129
ERROR	16	.0996		
<u>4.75 to 6.36 mm</u>				
TILLAGE	3	.0940	.75	0.5369
REP	1	.0013	.01	0.9199
TILLAGE * REP	3	.0874	.70	0.5659
ERROR	16	.1250		
<u>6.36 to 9.53 mm</u>				
TILLAGE	3	1313.1706	1.00	0.4197
REP	1	1322.8974	1.00	0.3313
TILLAGE * REP	3	1325.0339	1.01	0.4159
ERROR	16	1317.7344		

Table 28. ANOVA for the evaluation of the effect of glucose and nitrate on nitrous oxide production by the 2 to 4.75 mm aggregates of the Worthing soils.

## ANOVA

Dependent Variable = Nitrous oxide Production

for Glucose and Nitrate Addition to 2 - 4.75 mm Aggregates

<u>SOURCE</u>	<u>D.F.</u>	<u>Mean Square</u>	<u>F</u>	<u>PR &gt; F</u>
GLUCOSE	2	3082604	3.58	0.0492
NITRATE	2	1629185	1.89	0.1799
GLUCOSE * NITRATE	4	1048430	1.22	0.3385
ERROR	18	861992		